

K–T fossil patterns dispute abrupt mass extinction

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Nearly two decades ago appeared the remarkable hypothesis by Louis Alvarez and colleagues¹ according to which a large comet or asteroid (bolide) hit Earth at the end of Cretaceous and beginning of Tertiary (K–T boundary, about 65 Ma) which had resulted in wiping out of dinosaurs, ammonites and many other species living on the land and sea. Even though this impact event and the site of its impact over Mexico^{2,3} are generally accepted, a section of scientists differed with the views linking it with extinction of life; for them, the two were fortuitous coincidences. The prolonged debate that ensued on the impact–extinction dichotomy only created pro- and anti-impact-extinction groups, with neither side mustering enough evidence to convince the opposite group.

K–T extinction is not unique in Earth's long history and bouts of extinction, large and small, have appeared periodically; in fact, a 26 million year cycle of mass extinction episodes during the last 250

million years was proposed a few years ago by Raup and Sepkoski⁴. Palaeontologists have invariably linked these mass extinctions to terrestrial causes – geologic, climatic and ecologic. However, the K–T bolide hypothesis by the Alvarez team was notable for it added a new dimension to the on-going studies and set workers to seek extraterrestrial causes also to explain the decimation of biota. It had galvanized scientists around the world to rummage for extraterrestrial chemical links in the K–T strata and for impact-related repercussions on the biota, particularly those belonging to the sub-periods Maastrichtian and Danian lying on either side of this event horizon. Their search in several of the world's K–T sites yielded telltale enrichment of geochemically scarce Ir, as well as other elements like Se, As, Sb, Os, Ni; also, sudden changes in faunal diversity, steadily came to be recognized in many of these strata. Today, eighteen years after the K–T impact hypothesis was put forward, a mass of

literature and symposia proceedings on the subject have been published, but, the *en masse* death of dinosaurs and ammonites has remained as enigmatic and controversial as before.

Geological strata of K–T ages cover many parts of the globe, and well-preserved fossils of this age occur notably at Gubbio in Italy, El Kef in Tunisia, Seymour Island in Antarctica, Nye Kløv in Denmark, Caravaca, Zumaya, Agost in Spain, Brazos River (Texas), Colville River (Alaska), Montana and other places in USA, Majunga Basin in Madagascar, Mongolia, China, Peru and New Zealand (Figure 1). In India, well-known Maastrichtian–Danian equivalents occur in Tiruchirapalle and Pondicherry areas in southern India, Narmada River valley in western India, and in Meghalaya in the northeast India^{5–8}. Though several species of microbiota (nannoplanktons, Foraminifera), invertebrate (gastropods, bivalves, cephalopods, echinoderms) and vertebrate macrofauna (fish, reptiles,

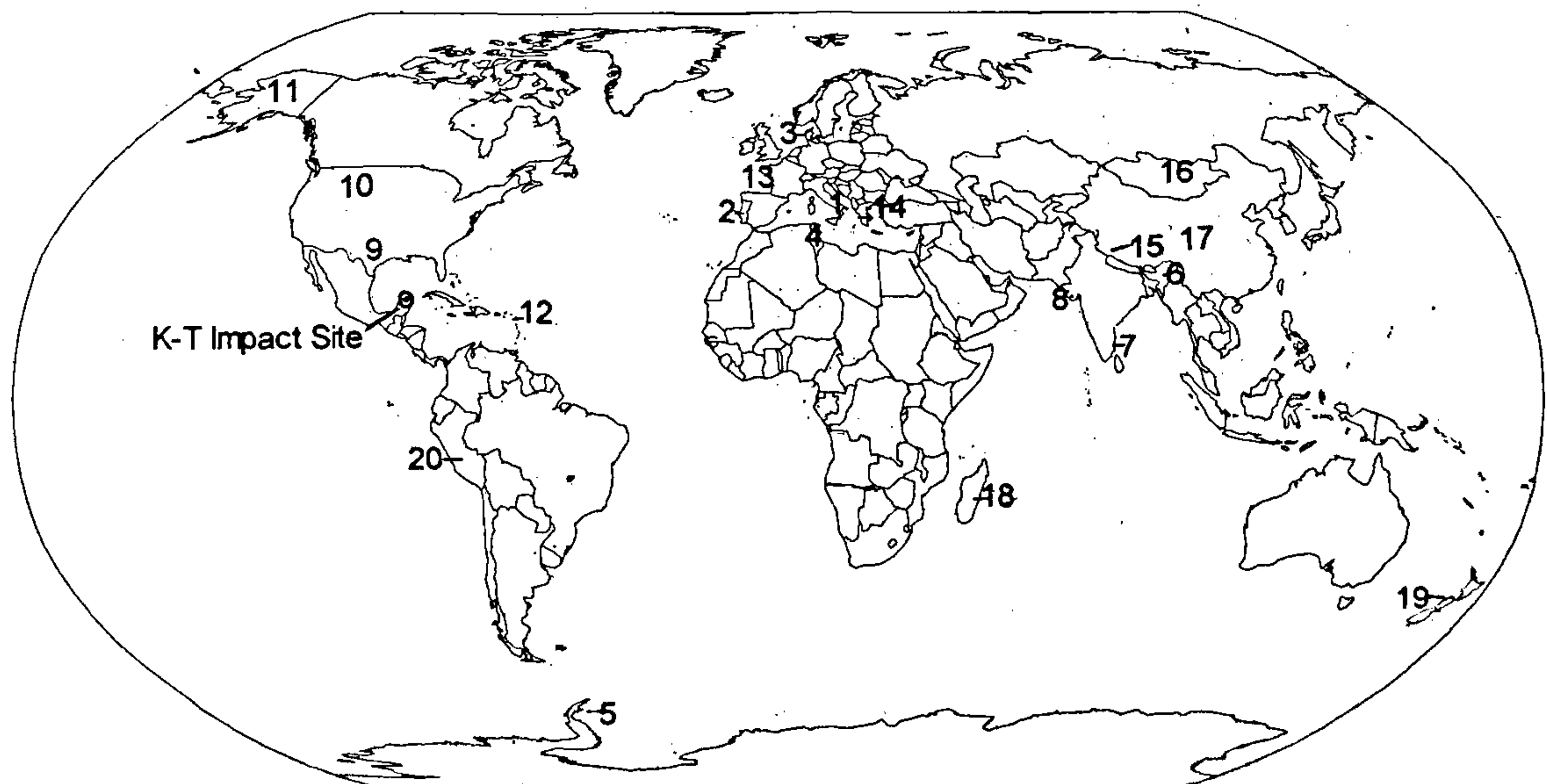


Figure 1. Cretaceous–Tertiary sites examined for fossil patterns: 1. Gubbio, Italy; 2. Caravaca, Zumaya, Agost, Spain; 3. Nye Kløv, Sterns Klint, Denmark; 4. El Kef, Tunisia; 5. Seymour Island, Antarctica; 6. Meghalaya, India; 7. Tiruchirapalle, India; 8. Narmada Valley, India; 9. Brazos River, Texas, USA; 10. Montana, USA; 11. Colville River, Alaska, USA; 12. Puerto Rico; 13. Bay of Biscay; 14. Bjala, Bulgaria; 15. Tethyan sites; 16. Mongolia; 17. China; 18. Majunga Basin, Madagascar; 19. Marlborough, New Zealand; 20. Peru, S. America.

RESEARCH NEWS

dinosaurs, amphibians) and flora have been reported from these Indian beds, their pattern of distribution in relation to the K-T event has been worked out only for a few localities—e.g. in the Um Sohryngkew river section in Meghalaya (N.E India), where faunal breaks, their decline/and or extinction/survival across K-T are documented^{7,8}.

Now, MacLeod (Museum of Natural History, London) and colleagues (University College and Birkbeck College,

London) have re-examined voluminous fossil records from a number of K-T sites around the world and have commented upon the environmental changes, their influence on life during that period and discussed the claimed abruptness of extinction⁹⁻¹¹ (Table 1). They have categorized the observed extinction patterns broadly as *progressive* (due to relatively long-term, moderate intensity environmental change), *catastrophic* (due to short-term moderate-high intensity epi-

sodes of environmental change) and *stepwise* (due to multiple short-term moderate-high intensity episodes of environmental change) and they view the K-T extinction patterns to be the outcome of combinations of these three mechanisms.

Secondly, in deciphering the life during K-T for which fossil patterns preserved in sedimentary rocks are much relied upon, MacLeod *et al.* have highlighted certain sedimentary processes that affect

Table 1. Summary of the state of K-T fossil record^{5,8,11}

Fossils	K-T Localities	Decline/extinction/emergence
Nannoplankton—Calcareous	Important marine boundary successions	10 species survived, 80 vanished.
Dinoflagellates	Tunisia, Antarctica	Pre-impact decline close to K-T due to sea-level rise.
Diatoms (unicellular algae having siliceous valves)	Seymour Island (Antarctica)	Survival across K-T 13–100% (84% for Antarctic species) attributed to dormant resting spores; unrelated to impact.
Radiolaria	USA, New Zealand	Occurrences patchy but continuity across K-T seen.
Planktonic foraminifera	Gubbio (Italy), Tunisia, Spain, Texas (USA), Meghalaya	Waves of extinction prior to, at and well after K-T due to eustatic sea-level/volcanic and impact-related events at or near the boundary.
Calcareous benthic foraminifera	South Atlantic, Tethyan sites, Spain, high southern latitudes, Tunisia	Opinion about extinction at K-T divided; ecological changes induced by eustatic sea-level shifts attributed.
Corals (reef-building and non-reef building types)	Tethyan habitat, Oman mountains	Extinction of reef-builders and their reemergence in Eocene; extinction related to collapse of algal symbiosis due to volcanic/impact/tectonic or climatic causes.
Bryozoans	Denmark, US Atlantic coastal plains, Madagascar	Abundant in late Cretaceous and Tertiary; data do not support abrupt K-T extinction.
Brachiopods	Denmark and a few other places in Europe	Sudden extinction at K-T boundary due either to facies change or to fluctuating climate; recovery in early Tertiary.
Cephalopods (<i>Nautilus</i> , <i>Ammonites</i> , <i>Belemnites</i>)	Denmark, France, Poland, Austria, Bulgaria, Georgia, Antarctica, India	Nautiloids continued across the K-T, ammonites and belemnites gradually declined preceding K-T and vanished at the event.
Gastropods and bivalves	Tethyan sites, Arabia, India, N. France, Baluchistan, Puerto Rico, Madagascar	Most gastropods, bivalves declined and vanished by Cretaceous end, only a few survived into Tertiary; some bivalves showed diversity reduction at the K-T boundary.
Echinoderms (<i>Holothurids</i> , <i>Asteroids</i> , <i>Crinoids</i> , <i>ophiuroids</i> , <i>echinoids</i>)	Denmark, northern Europe, NW Europe, Kazakstan	<i>Holothurian</i> records poor; <i>Ophiuroids</i> and <i>Asteroids</i> survive K-T; <i>Crinoids</i> decline after Palaeocene; <i>Echinoids</i> decline across K-T due to loss of habitat and facies changes.
Fishes	600 families, over a wide range of sediment types around the world	35 cartilaginous families survived K-T and 7 became extinct; reduced survival rate of many others not impact related.
Amphibians	North America, S. America, Europe, Africa, Asia, S.E. Asia, India, Sudan, Seychelles	No amphibian extinction at or near K-T boundary and several families survive K-T event unaffected.
Lepidosaurs (<i>Rhyncocephalia</i> , <i>Squamata</i> , <i>Mosasaurus</i>) land and marine reptiles	N. America, China, Mongolia, New Zealand, India	<i>Rhyncocephalia</i> decline by mid-Cretaceous; <i>Squamata</i> , survived K-T; <i>Mosasaurus</i> , marine lizards decline and fail to survive K-T due to eco-changes.
Archosaurs (dinosaurs, pterosaurs, crocodiles and birds)	Bolivia, North & South Americas, West Africa, Antarctica, India, China, Morocco	50% of <i>Crocodylians</i> survived K-T; pterosaurs and some dinosaurs decline at K-T, rates varying geographically; Bird ancestors, except shore-birds, extinct at K-T; many bird orders branched surviving K-T.
Terrestrial plants	Western USA, Antarctica, Canada, New Zealand	Megafloral changes at/after regional K-T boundary; microflora in US sites show abrupt extinction at K-T.

their pattern, which, if overlooked, may lead to wrong conclusions. For example, changes in the rates of sedimentation may appear as if there had been an abrupt or gradual cessation of deposition (hiatus); and likewise, changes in sedimentary material or their composition may provide favourable conditions for excellent fossil preservation. These aberrations which sway distribution patterns, if ignored, may bias conclusions regarding species diversity, their decline, extinction or survival. The global decline of ammonites during early to late Maastrichtian falls in this category as this pattern may be due to sampling bias resulting from a reduction in the number of known localities rather than a reflection of the biotic pattern. Thirdly, the migration of biota due to ecological changes, a feature frequently noticed among both terrestrial and marine species (sharks are known to have migrated to distant geographic sectors due to sea-level changes)¹², or reworking of older strata leading to transport of fossils to younger horizons may, falsely, suggest an extended range, the former spatially, and the latter, temporally. An example of transfer of fossils is the reworking seen in south Indian Tiruchirapalle Cretaceous site where Maastrichtian fossils of the older Ariyalur and Niniyur Groups have been transported into younger Tertiary horizon¹³. Fourthly, index fossils, though good as guides for dating, cannot be relied upon as unambiguous datum markers, e.g. ammonites or dinosaurs, though typically Mesozoic, may occur in younger beds also. Lastly, patchy or poor fossil preservation, phylogenetic and taxonomic blunders, and errors in sampling and laboratory procedures are other pitfalls that are known to have affected K-T fossil studies in the past.

MacLeod *et al.* feel that the late Cretaceous period, preceding the K-T, witnessed combinations of earth-bound events (global climatic changes induced by extensive volcanisms or eustatic sea-level changes) which disturbed to varying degrees biotic groups in diverse geographic habitats, leading to decline in their diversity. The much-publicized decline of groups of marine calcareous nannoplankton and planktonic foraminifera close to the K-T boundary, according to them, is unrelated to bolide impact since this trend is noticed even prior to the impact. Some of these and dinoflagellate groups are seen surviving into

younger Danian strata, but whether this is due to species survival or the result of reworking of components of older Cretaceous fauna, is debatable. Isotopic tests, however, support the former interpretation. In India too, at the Meghalaya site, the planktonic foraminifera and nannoplanktons seem to disappear suddenly at K-T, but their continuation well above this boundary suggests that instantaneous extinction does not apply to them⁸. Similarly, the pattern exhibited by various other forms of life on land and sea shows decline/or extinction prior to the K-T event, a few at the K-T while some are observed to survive across this boundary (see Table 1). Whatever be the reasons for species decline and survival, the faunal diversity took a long time to recover and the rates of their recovery also varied geographically—the North American marine bivalves taking longer time than those from Europe or Asia. Lastly, one major aspect that has frequently clouded precise interpretation of the K-T scenario has been the doubtful judgment of phylogenetic relationship (species and genus levels) between Cretaceous and Danian taxa.

MacLeod and others are skeptical about the magnitude and abruptness of extinction within a very narrow time interval at the very end of Cretaceous. Instead, they maintain that the adverse climatic and ecological changes were pulsating throughout the upper Cretaceous with which the faunal fluctuations observed are correlatable. There is also a view that such wave-like fossil patterns (stepwise extinction and survival) may be the result of short-term (few million years) cometary impacts¹⁴. In another radical approach to explain this wavelike extinction patterns, Richard Sole (Universitat Politecnica de Catalunya, Barcelona, Spain) and colleagues identify a fractal structure in the fossil record, which is 'consistent with statistical measures such as frequency distribution of extinctions and lifetimes, which also exhibit power-law decay'¹⁵. They apply complexity theory to explain mass extinctions, where a normally stable biota, through interactions of the components within the system, get poised in a state of 'self-organized criticality'¹⁶—a delicate state during which a minor climatic change will precipitate cascading effects that may wipe out most of the species. Sole *et al.* conclude that the extent of response to a major mass ex-

inction event is governed by internal biotic organization and not the magnitude of external perturbations. The extinction will therefore be fluctuating, following the power law distribution—a feature which according to the authors is well applicable to the K-T fossil patterns.

David Jablonski (University of Chicago) feels that it is indeed difficult to differentiate the 'effects of sudden event from those of larger trend' like, for example, drop in sea-level which will not only reduce habitable area in marine shelves and swamps leading to extinction but also 'reduce outcrops or the breadth of palaeoenvironments available for sampling'¹⁷. Notwithstanding the maze of interpretative hazards in assessing the K-T extinction, and putting aside judgement on the mountain of evidences upholding impact triggered mass extinction, undoubtedly any answer to this raging controversy^{18,19} has to be supported by palaeontological records. Current interdisciplinary efforts employing improved isotopic dating, proper verification of temporally displaced groups, precise species identification and statistically verified quantitative portrayal, are bound to provide answers as to how much of the biotic ups and downs of late Mesozoic-early Tertiary times are influenced by extraterrestrial events.

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NEWS

Niels Bohr-UNESCO Gold Medal

Professor S. Chandrasekhar, Centre for Liquid Crystal Research, Bangalore has been nominated by the Niels Bohr Institute for the award of a Niels Bohr-UNESCO Gold Medal in recognition of his 'outstanding contributions to the development of liquid crystals and the

advancement of science in developing countries'. The presentation will be made by the Director General of UNESCO, Dr Federico Mayor, during the symposium on 'Niels Bohr and the Evolution of Physics in the 20th Century' to be held in Paris 27-29 May 1998.

Three other distinguished physicists will also be receiving the NBU Gold Medals on this occasion: Professor V. L. Ginsburg, Russia (who is an Honorary Fellow of the Indian Academy of Sciences and of INSA), Professor Walter Kohn, USA, and Professor Alexander Polyakov, USA.

SCIENTIFIC CORRESPONDENCE

An improved method for the isolation of supercoiled plasmid DNA

Rapid methods of small-scale preparation of plasmid DNA are very common in molecular biology and genetic engineering. Several procedures are in vogue at present to produce plasmid DNA suitable for various molecular biology experiments. Mini-preparations of plasmid DNA by alkaline lysis¹ and boiling² methods contain chromosomal DNA and require cumbersome phenol, chloroform extractions. Most of the commercial kits that are available for mini-preparation of pure plasmid DNA are very expensive.

Here we describe a simple inexpensive procedure for mini-preparation of supercoiled plasmid DNA suitable for various molecular biology techniques. This method is an adaptation of alkaline lysis method³. The modifications include the use of MgCl₂ to remove the RNA⁴ and the use of 1 M sodium acetate instead of 3 M as used in the original alkaline lysis method, to get rid of chromosomal DNA contamination totally. The modified method is simple, needs only bench top chemicals and a microfuge and does not include any costly chemicals such as caesium chloride. Further, in the modified method the inconvenient phenol, chloroform extraction steps are also avoided. The entire operation of the procedure takes less than 1 h and the yield is

approximately 3 to 4 µg per 1.5 ml *E. coli* culture.

A 5 ml overnight culture of *E. coli* strain containing specific plasmid is grown in LB containing the required antibiotic. About 1.5 ml of the culture is spun in a microfuge tube at 10,000 rpm for 2 min at room temperature. The pellet is suspended in 100 µl TEM buffer (50 mM Tris-HCl, pH 8.0, 10 mM EDTA and 100 mM MgCl₂) and to this suspension 200 µl of freshly prepared NaOH/SDS solution (0.2 N NaOH and 1% SDS) is added, with gentle mixing by inverting the tubes. The tube is kept on ice for 5 min. To the above, 150 µl of 1 M sodium acetate pH 4.6 is added and mixed gently by inverting the tube. Vortexing of the tube is totally avoided. After 5 min of incubation again on ice, the tube is spun for 5 min at 10,000 rpm at room temperature. The supernatant is transferred to a fresh tube, mixed with 280 µl isopropanol and placed on ice for 5 min. The pellet is collected by centrifugation at 10,000 rpm for 5 min, washed with 70% alcohol and air-dried briefly. It is important not to let the pellet dry completely.

The pellet is resuspended in 100 µl 1 M sodium acetate, pH 4.6 and incubated on ice for 5 min. The suspension is then

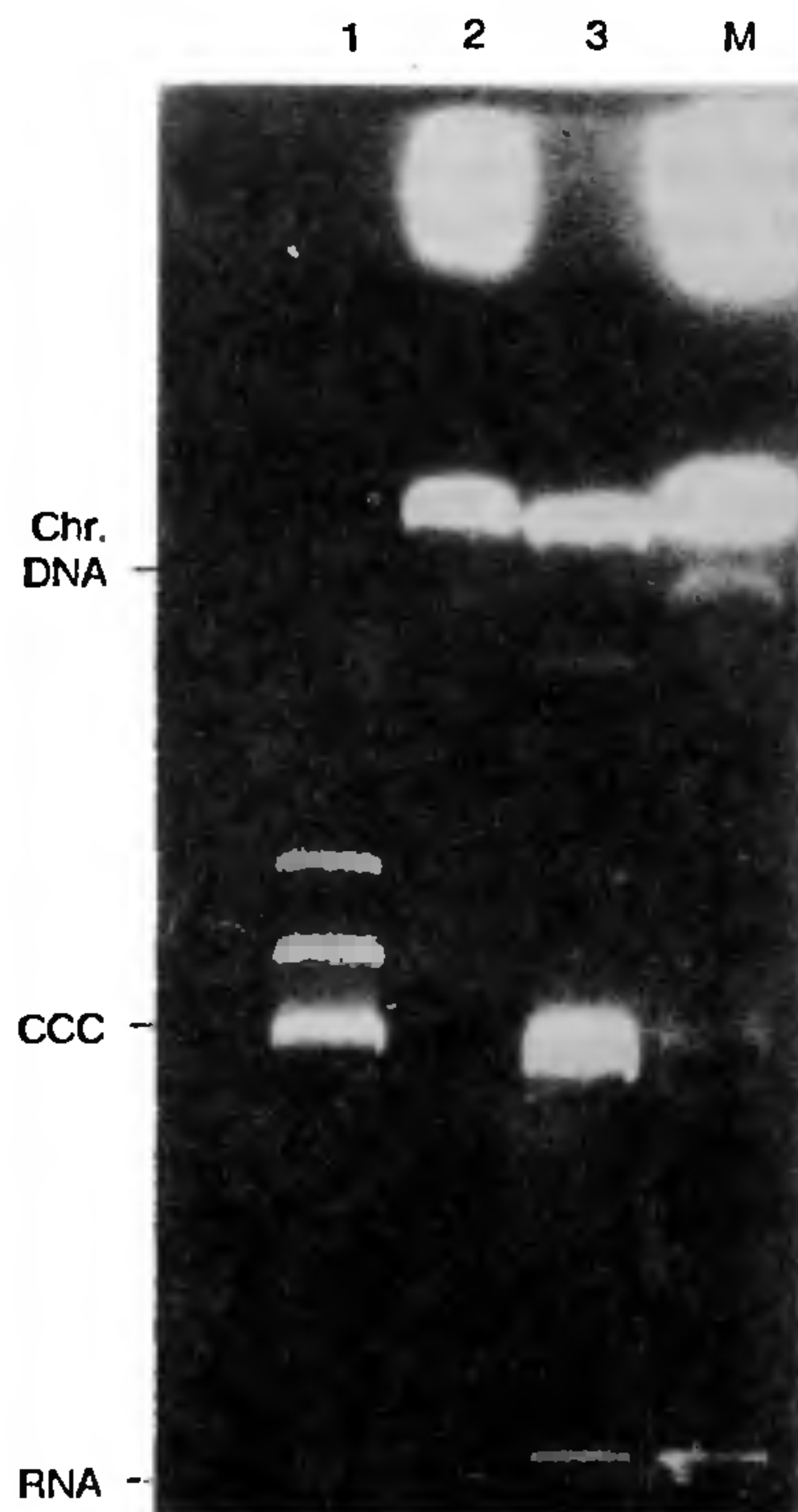


Figure 1. Mini-preparations of pUC 18 plasmid DNA. Plasmids isolated from 1.5 ml culture following different methods were suspended in 30 µl TE and 5 µl from each isolate was analysed on 0.7% agarose gel. The lane marked M contains *Hind*III cleaved λ DNA as molecular weight markers. Lane 1, Alkaline lysis method; Lane 2, Boiling method; Lane 3, Method described in this paper.