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22. TRIPOS Associates, Inc., 1699 South Hanley Road, Suite 303, St. Louis, MO 63144.
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24. Spectroscopic data for *racemic*-$Rh_2(\mu\text{-}CO)_2(CO)_2$(et,ph-P4): dark red oil; infrared spectra ν_{CO}: 1985 cm^{-1} (sh), 1957 cm^{-1} (s), 1813 cm^{-1} (sh), 1768 cm^{-1} (m); $^{31}P\{^1H\}$ NMR (40.48 MHz, acetone-d_6, versus 85% H_3PO_4): δ 71.6 (dt, 1P, $J_{P\text{-}Rh}$ = 96 Hz and $J_{P\text{-}P}$ = 23 Hz), 26.0 (dt, 1P, $J_{P\text{-}Rh}$ = 96 Hz and $J_{P\text{-}P}$ = 23 Hz).
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Paleoatmospheric Signatures in Neogene Fossil Leaves

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An increase in the atmospheric carbon dioxide (CO_2) concentration results in a decrease in the number of leaf stomata. This relation is known both from historical observations of vegetation over the past 200 years and from experimental manipulations of microenvironments. Evidence from stomatal frequencies of fossil *Quercus petraea* leaves indicates that this relation can be applied as a bioindicator for changes in paleoatmospheric CO_2 concentrations during the last 10 million years. The data suggest that late Neogene CO_2 concentrations fluctuated between about 280 and 370 parts per million by volume.

Analysis of air locked in polar ice has been used to document the paleoatmospheric CO_2 concentration for the past 160,000 years (*1*). Concentrations of CO_2 from before this time cannot be directly measured and can only be inferred from forward modeling of the CO_2 global budget (*2*) or from proxy records that are based on geochemical or paleontological observations. The geochemical approach depends on the interpretation of changing $^{13}C/^{12}C$ isotope ratios in the marine carbonate record (*3*) and, more recently, on the analysis of $^{13}C/^{12}C$ ratios of soil carbonates (*4*). A relevant paleontological approach is highly dependent on the ecophysiological interpretation of the morphological or anatomical characteristics of fossils (*5*). Because of the prominent role of photosynthetic CO_2 fixation in the interaction between the atmosphere and biota through time, land plants represent one of the most obvious categories of organisms to be investigated for paleoatmospheric signatures. In this report, we illustrate that the analysis of stomatal frequencies on fossil leaves may be used to determine CO_2 concentrations during the last 10 million years.

Physical models of CO_2 fixation in land plants with C_3 metabolism (*6*) can be used to show that the areal density of leaf stomata is responsive to changes in ambient CO_2 concentration. Under nonlimiting light and water conditions, leaves have an optimal CO_2 fixation rate at a stomatal conductance to diffusion of CO_2 at which the CO_2 concentration is maintained in the leaf interior at the break point between saturation and limitation of the CO_2 receptor molecule ribulose biphosphate (RuP_2). The stomatal conductance to diffusion is a complex parameter that is dominated by the density and the aperture size of the stomata. The maintenance of optimal CO_2 fixation may require that an increase of atmospheric CO_2 concentration be accompanied by a lower value of the stomatal conductance. Hence, if we assume that differences in stomatal frequency are part of the phenotypic plasticity of a plant species, such a lowering could be reached through a decline of stomatal density (*7*).

This possibility is supported by the results from the study of herbarium material collected over the last 200 years. Research by Woodward (*8*) emphasized that the human-induced CO_2 increase has resulted in a mean reduction of 40% in the stomatal density of European temperate forest trees. The significant decline was confirmed by experiments under controlled ambient CO_2 concentrations (*9*) and could be calibrated against historical CO_2 concentrations inferred from ice cores. An important corollary of this stomatal response to changing CO_2 regimes is that analysis of stomatal frequencies on fossil representatives of extant plant species could help to determine paleoatmospheric CO_2 concentrations at different geologic times. Because of the fossilization potential of the biopolymer cutan (*10*), analysis of fossil leaf cuticles can provide relevant information on stomatal frequencies. Several studies have explored the potential of cuticle analysis in late Pleistocene and Holocene paleoatmospheric research (*11*). In this report, we concentrate on the late Neogene. We based our data on the study of the cuticle of *Quercus petraea* (Durmast oak) from successive time intervals that were characterized by contrasting climatic conditions.

The species *Q. petraea* is essentially European, with a present distribution from southern Scandinavia to the Mediterranean region. The fossil record of this deciduous oak extends back to the late Miocene (*12*). We analyzed leaf compressions (Fig. 1) that were collected from clay intercalations in terrestrial late Miocene and Pliocene sequences of the Lower Rhine Embayment (the current southeastern margin of the Neogene North Sea Basin exposed in Germany and adjacent parts of the Netherlands). Source strata were deposited during five successive, regionally recognized time intervals between ~10 million years ago (Ma) and ~2.5 Ma (*13*), namely, base

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Linne B and C, top Linne B and C, Susterian, early Brunssumian, and late Reuverian (Fig. 2).

Well-preserved abaxial leaf cuticles of fossil *Q. petraea* reflect the epidermis cell structure and stomatal arrangement. Thus, we were able to count the stomatal frequency as a percentage of the total number of epidermal cells [stomatal index (*14*)]. Because of the generally limited material available, a direct interpretation in fossil studies of stomatal densities in terms of paleoatmospheric change may give unreliable results. In individual species of living plants, the number of stomata per unit area is known to vary significantly in response to environmental factors other than the ambient CO_2 concentration. A decreased stomatal density may be the result of decreasing light intensity or increasing humidity. However, because such changes coincide with a reduction in the number of epidermal cells, in contrast to the stomatal density, the stomatal index represents a satisfactory constant for any one plant. Experiments have shown the stomatal index as generally reliable except under extreme environmental stress (*15*). Therefore, if sufficiently well-preserved cuticles are available in fossil studies, reliable stomatal indices can be calculated on the basis of a series of measurements on a limited number of leaves.

Fig. 1. Fossil of *Q. petraea* from the Lower Pliocene of the Lower Rhine Embayment. Magnification, ×0.4.

Stomatal indices calculated for fossil *Q. petraea* from the Linne B and C, early Brunssumian, and late Reuverian intervals show mean values between 9.5 and 11.5 (Fig. 2). In contrast, the mean value of 16.2 for leaves from the Susterian is significantly higher. To test the validity of this difference, we analyzed cuticles of an extinct species, *Fagus attenuata*, from both the Susterian and top Linne B and C intervals. Again, the stomatal index for Susterian leaves (14.1 ± 0.5) exhibited a significant increase relative to the values for Linne B and C material (10.0 ± 0.8).

The changes in the stomatal index for *Q. petraea* suggest a covariation with temperature-related climatic shifts that have been inferred from the late Miocene-Pliocene palynological record of the Lower Rhine Embayment (Fig. 2). Comparison with the extensive record of plant megafossils, notably those of fruits and seeds (*16*), confirms that changes in quantitative composition of pollen assemblages (*13*) reflect a broad-scale vegetation change characterized by an alternation of mixed evergreen-deciduous and deciduous forest types. On the basis of these fossil data, warm-temperate to subtropical conditions are thought to have prevailed during the Linne B and C intervals, with a temperate climate during the Susterian, and warm-temperate conditions in early Brunssumian and late Reuverian times. The differences in the thermal regimes constrain the distribution of modern Northern Hemisphere analogs for the two contrasting forest types (*17*). Thus, the late Miocene-Pliocene vegetation changes suggest fluctuations of 2° to 3°C in the regional mean annual temperature.

In addition to analyzing fossil material, we calculated stomatal indices that cover the last 120 years for herbarium specimens of *Q. petraea* (Fig. 3). The observed decline in mean values with time corroborates Woodward's (*8*) conclusion that the anthropogenically induced global CO_2 increase has resulted in a proportionate decrease in stomatal frequencies for forest trees (*18*). Present-day stomatal indices for *Q. petraea* are similar to those for fossil material from the Linne B and C, early Brunssumian, and late Reuverian intervals. On the other hand, indices for times before the development of large-scale industrialization appear to be comparable to those of the Susterian record.

The recorded changes in the stomatal

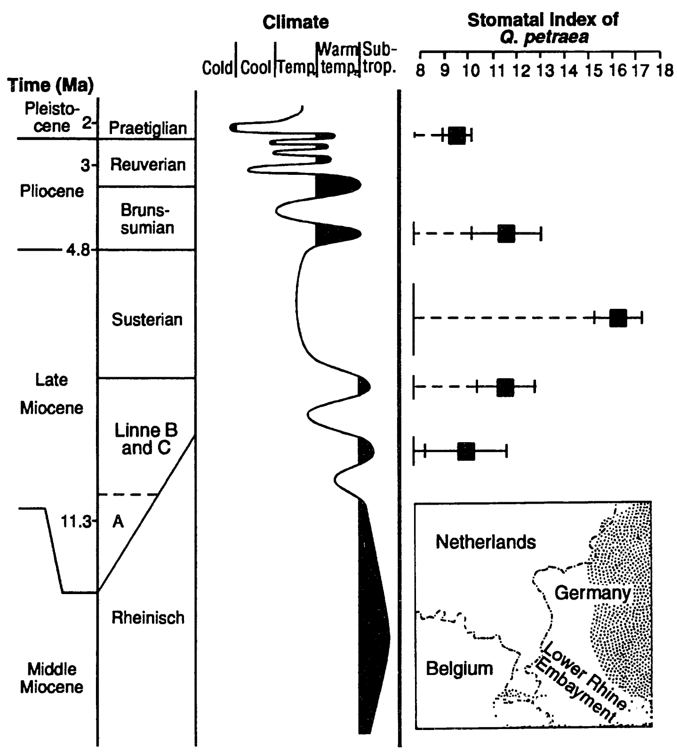

Fig. 2. Stomatal indices for fossil *Q. petraea* compared with major Neogene climatic oscillations inferred from the late Miocene-Pliocene pollen record in the Lower Rhine Embayment. The stomatal index is a measure of the stomatal frequency as a percentage of the total number of epidermal cells (*14*). Regional stratigraphy and the climatic curve are modeled after the work of Zagwijn and Hager in (*13*). Vertical bars are estimated stratigraphic uncertainties of the investigated leaf floras. Data points are mean ± SD. Temp., temperate; Subtrop., subtropical.

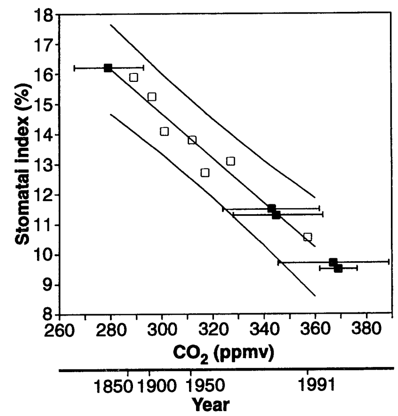

Fig. 3. Stomatal indices for Neogene *Q. petraea* compared with the historical relation between atmospheric CO_2 concentration and stomatal indices for *Q. petraea* during the last 120 years. Recent stomatal indices (□) have been calculated from cuticles of herbarium material collected in western Europe between 1873 and 1991. Historical increases of mean global CO_2 concentration correspond to data from Antarctic ice cores and direct measurements at Mauna Loa, Hawaii (*19*). The linear regression line, with 95% confidence limits, shows a reduction of stomatal indices over the studied period; slope, −0.074 ± 0.008. Neogene stomatal indices (■) plotted on the regression line are means for five time intervals (compare with Fig. 2). Horizontal bars indicate the inferred ranges of paleoatmospheric CO_2 concentration.

index for an individual plant species confirm the use of stomatal frequencies as proxy indicators of fluctuations in paleoatmospheric CO_2 concentration. Calibration of the stomatal indices against the historical relation between stomatal frequency and CO_2 concentrations (*19*) enables quantification of the late Miocene-Pliocene paleoatmospheric signals (Fig. 3). The relative changes so far observed suggest that the corresponding global CO_2 concentration has fluctuated between values of ~280 and ~370 parts per million by volume (ppmv). Covariation with climatic changes supports a causal relation between the CO_2 regime and temperature in late Miocene to Pliocene times. On the basis of such a relation, present-day low stomatal indices suggest that the presumed climatic effects of the human-induced CO_2 increase are lagging behind the stomatal responses in land plants.

The fossil leaf record is characterized by its generally discontinuous nature. The relatively few samples available in any one sedimentary sequence limit the extent to which detailed patterns of paleoatmospheric change can be reconstructed. However, quantitative stomatal analysis of cuticular remains can be used to test whether regional relative temperatures that are inferred from more continuous palynological records consistently reflect paleoatmospheric change (*20*).

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$$A = g_c(c_a - c_i)$$

where A is the assimilation rate, g_c is the conductance to diffusion of CO_2 (the reciprocal of resistance), and c_a and c_i denote CO_2 concentrations in the atmosphere and in the intercellular leaf spaces, respectively. A commonly recognized effect of rising c_a is a decrease in g_c; see S. P. Long, N. R. Baker, C. A. Raines, *Vegetatio* **104/105**, 33 (1993).
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Recent Variability in the Southern Oscillation: Isotopic Results from a Tarawa Atoll Coral

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In the western tropical Pacific, the interannual migration of the Indonesian Low convective system causes changes in rainfall that dominate the regional signature of the El Niño–Southern Oscillation (ENSO) system. A 96-year oxygen isotope record from a Tarawa Atoll coral (1°N, 172°E) reflects regional convective activity through rainfall-induced salinity changes. This monthly resolution record spans twice the length of the local climatological record and provides a history of ENSO variability comparable in quality with those derived from instrumental climate data. Comparison of this coral record with a historical chronology of El Niño events indicates that climate anomalies in coastal South America are occasionally decoupled from Pacific-wide ENSO extremes. Spectral analysis suggests that the distribution of variance in this record has shifted among annual to interannual periods during the present century, concurrent with observed changes in the strength of the Southern Oscillation.

The ENSO system of the tropical Pacific governs interannual variability throughout the tropics and imparts its signature to climate worldwide (*1*). In the western Pacific, ENSO-related changes in the location and intensity of the Indonesian Low convective maximum lead to dramatic shifts in precipitation patterns. At Tarawa Atoll, measurements indicate that intense rainfall alters the salinity of the underlying surface ocean by up to 4 per mil (*2*) and that surface water $\delta^{18}O$ (*3*) varies linearly with salinity. These isotopic changes provide the means by which we can trace the past migrations of the Indonesian Low beyond the historical record with the use of high-

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