

Fig. 2. (A) Laboratory arrangement (at left) showing the compactness of the device; a 6-inch (15-cm) ruler is shown beside the pulse unit. The specimen location is at the far right. (B) Time behavior of beam-discharge operation; the horizontal axis is 100 nsec per division. (Upper trace) Beam current at 2 kA per vertical division; (lower trace) beam current plus discharge current at 3 kA per vertical division. (C) Soft x-ray replica of a diatom as seen in optical microscopy by using a Leitz Orthoplan with interference contrast. (D) High-resolution sample viewing with a Hitachi S-500 scanning electron microscope. Scale bars in (C) and (D) are labeled in micrometers.

storage, both stages being chargeable from a common d-c supply. The time behavior of the operating device is shown in Fig. 2B; the beam current pulse ( $\sim 60$  nsec at full width at half-maximum) is shown on the upper trace and the beam current plus the discharge current is shown on the lower trace. X-ray pulse duration is less than the total current duration of  $\sim 100$  nsec.

Single-shot exposures of objects placed directly on the resist surface were obtained (see Fig. 1B). The x-ray resist, polymethyl methacrylate, was spun onto the surface of a silicon wafer for mounting in the vacuum chamber. Development was in a 1:1 solution of methyl isobutyl ketone and isopropanol, development time being 3 to 6 minutes. Exposures were made at a distance of 22 cm from the output end of the flash x-ray tube, with the center of the wafer on axis with the tube. Penumbra blurring was thus less than  $1 \text{ \AA}$ . Developed resists can be viewed with a high-resolution optical microscope, from which structures  $\sim 0.5 \mu\text{m}$  or greater in size are apparent (see Fig. 2C). Higher resolution observations were made by scanning electron microscopy following evaporation of a  $50\text{-\AA}$  palladium-gold overcoat (Fig. 2D). A resolution of  $\sim 300 \text{ \AA}$  was realized.

The spectral emission from the flash tube in the range useful for wet-sample viewing is indicated by a densitometer tracing in Fig. 1C. The spectrum was recorded on a 2.2-m grazing incidence vac-

uum spectrometer (McPherson model 247) with a platinum grating (300 lines per millimeter) and SWR (short-wavelength radiation) plates. Also shown in Fig. 1C are the absorption coefficients for water, protein, and the excess of protein over water. (Note that the curve for

protein minus water is a strict subtraction of absorption coefficients, direct use of which in the exponential would imply equal path lengths in both materials.) The coincidence of the spectra with the wet-specimen window and their distribution within the window are such as to provide optimum contrast and maximum resolution ( $l, 3$ ). Thus, this source should be quite useful for wet-sample viewing.

R. McCORKLE  
J. ANGILELLO, G. COLEMAN  
R. FEDER, S. J. LA PLACA  
IBM Thomas J. Watson  
Research Center,  
Yorktown Heights, New York 10598

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6. The expert efforts of F. Babbage and B. Kahn in device fabrication and of C. Aliotta in SEM viewing and the helpful comments of D. Sayre are gratefully acknowledged.

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## Rainfall Changes in Summer Caused by St. Louis

**Abstract.** *Precipitation in and around St. Louis was investigated to study urban influences on summer precipitation conditions. Prerain winds were used to define the "downwind area" where influences would be greatest, and wind-sorted rains were combined into monthly and summer totals. Seventy-five percent of the 16 rain patterns revealed a rainfall maximization downwind of the city, and the rainfall in the downwind area was 22.7 percent more than the rainfall upwind of St. Louis where no urban influences existed. Various statistical tests of the summer rainfall distribution reveal the downwind rainfall to be significantly greater than elsewhere and supportive of other findings that St. Louis increases rainfall.*

Climatological research results of the past 20 years (1) have indicated that major urban areas influence clouds and precipitation. Sizable changes,  $> 10$  percent, have been considered controversial (2). A major 5-year meteorological project was launched at St. Louis in 1971 to study intensively how an urban area modifies the atmosphere, how physical processes in clouds and rainfall are subsequently changed, and where any anomalous precipitation occurs (3). This experiment has provided a wealth of weather data and a variety of results that collectively indicated sizable ( $> 10$  per-

cent) localized increases in summer rainfall and storminess (4). It is difficult to evaluate inadvertent urban modification of rain because there is no randomization, and thus a "data analysis" approach that combines physical insight and statistical tests appears well suited (5).

A potentially definitive, yet simple physical-statistical test of the apparent urban-related rainfall increase at St. Louis is based on the determination of the placement and magnitude of maximum rainfall areas near St. Louis under differing wind directions. The basic hy-

pothesis is that the low-level winds moving across the urban area define a plume of urban-altered air (aerosols, heat, and moisture) that can affect rainfall over and beyond or downwind of the city (6). Precipitation elements move in all directions, often not in the same direction as the low-level winds, but most precipitation elements in the St. Louis area move from westerly directions and therefore one would expect the greatest influence to be exerted essentially east of the city (7).

The urban plume and the precipitation elements are known to have dual and often dissimilar motions. Nevertheless, if urban influences are real, a rain increase downwind (based on the low-level wind or plume direction and not the precipitation motion) of the city should presumably occur under any low-level wind direction. Hence, a maximum should be located west of St. Louis when winds are from the east. To this end, I compared rainfall values downwind of the city, under different wind directions existing before each rain began, with those upwind and on either side of St. Louis.

Rainfall was collected at 220 recording rain gages evenly distributed within a circle 80 km in diameter centered on the city (Fig. 1). Studies of the rainfall revealed 302 individual rainfall events in the five summers from 1971 through 1975 (8). For each of these rain events, the low-level wind direction during the 3 hours prior to the rain was determined from available wind data at 28 stations coupled with an analysis of local synoptic weather conditions. Thus, the prevailing winds before the rain were used to provide a general estimate of the urban plume or area of influence. Whether the urban influence is related to temperature, moisture, aerosols, or all three factors is unimportant here; the plume serves only as a means of defining downwind areas of influence on the atmosphere and upwind (control) areas of no influence.

The circular network was divided into four equal-sized quadrants (Fig. 1). The prerin winds were sorted into four directions of motion: northwest, northeast, southeast, and southwest. Then, for each direction, the quadrant rainfall was summed by the month and the summer.

The four direction patterns for June revealed that the downwind quadrant had the maximum in three of four cases (Table 1); in the case of southeast pre-storm winds, the high was in the quadrant to the right of the city. In July the highest quadrant value occurred down-

Table 1. Placement of maximum quadrant rainfall with respect to St. Louis and for each month. The maximum could be downwind, upwind, left, or right of St. Louis.

Prerin wind direction	June	July	August
North-west	Down-wind	Down-wind	Down-wind
South-west	Down-wind	Down-wind	Down-wind
South-east	Right	Down-wind	Right
North-east	Down-wind	Down-wind	Right

wind for all four directions. In August, two of the four directions (southwest and northwest), had the maximum in the predicted downwind area. Thus, in 9 out of the 12 possible monthly cases, the maximum occurred in the downwind location. The high was always downwind of St. Louis in all months (Table 1) when southwest and northwest prerin winds occurred. These are also the directions of the predominate motions of individual summer rain elements. These results indicate that the urban influence was great-

est when the low-level winds and storm cell motions were aligned (9). When northeast and southeast winds occurred and when the three monthly reversals occurred (maximum not downwind), the urban plume was moving generally in a direction opposite to the approaching storm motion, interacting with the rain well before reaching the city or downwind area, and hence being diffused and realized in the areas to the left or right of the city.

Figure 1A presents four maps showing the rain values with each wind direction for the five summers (June through August) sampled, 1971 through 1975. The southeast, the southwest, and the northwest prerin wind conditions have their maximums downwind. However, the northeast pre-storm winds have a maximum to the right of the city.

Figure 1B shows the total summer rain pattern derived if the four wind patterns are combined with respect to a common wind direction. The values demonstrate the marked downwind maximization. The difference between the downwind value of 129.4 cm and the upwind value of 105.5 cm is 23.9 cm, and this represents a 22.7 percent increase in rainfall in the area downwind of the city, regardless of wind direction.

Three statistical tests were applied to the quadrant rainfall values to examine the claim that there is no urban effect on precipitation. In the first test I examined the frequency of maximum rainfall in the downwind area, in the second test the magnitude of the downwind rainfall (versus the rainfall elsewhere), and in the third test the areal distribution of the ranks achieved by the values of the four areas. The only assumption in each test was that the events tested were independent, which is highly likely when one considers that these were monthly values sorted on the basis of wind directions.

In the first test, each of the 12 monthly outcomes was viewed as an independent Bernoulli trial. The null hypothesis of the probability  $P$  being equal to one-fourth was tested; that is, the expected distribution of maximum values was one in four, versus the hypothesis that the downwind distribution was greater than one in four. Thus, if there is no urban effect or no association between prerin winds, the city, and the downwind rainfall, there is a 25 percent likelihood of having the high in the correct (downwind) position. In the sample obtained, the downwind frequency was nine maxima, which was found to be significant at  $P > .0004$ . Thus, the hypothesis that the wind cross-

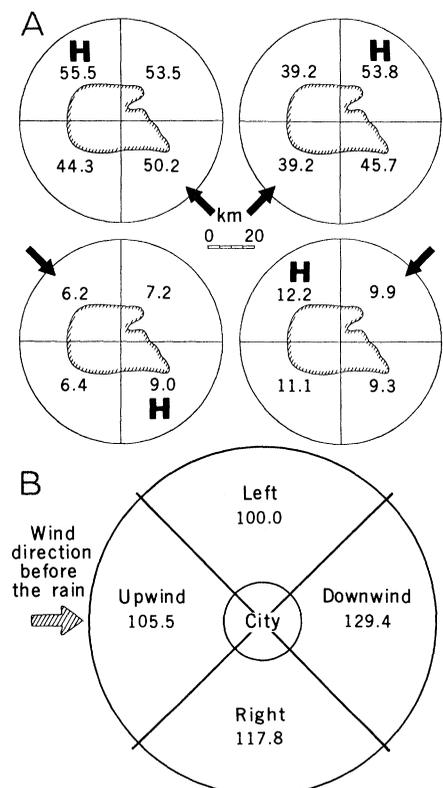


Fig. 1. (A) Four patterns of total summer rainfall (in centimeters) in quadrants around St. Louis, each associated with one of four prerin wind directions during the period 1971 through 1975. (B) A composite of the total summer rainfall (1971 through 1975) (in centimeters) positioned with respect to the prerin winds.

ing the city plays no role in the maximization of the downwind rainfall cannot be sustained. The maximum occurred downwind with sufficient frequency to be declared highly significant, if one uses the binomial test.

In the second statistical test, I evaluated the quadrant rainfall values in the 12 monthly wind-rain patterns by comparing the downwind values with those in the other quadrants. The differences, say, for downwind versus upwind, were ranked, and the one-sample Wilcoxon test was applied to these ranks. Addition of the negative ranks (upwind > downwind) for each comparison provided a number tested for its significance. The downwind versus upwind differences were significant at  $P < .013$  (10). This comparison indicated that the downwind rainfall value differed greatly from those of the three other areas around St. Louis.

In the third test I used the ranks of the quadrant rainfall values in each summer month for each wind direction. For each of the 12 patterns, ranks were assigned with the maximum quadrant given rank 1 and the minimum given rank 4. These were summed to get a summer score for each quadrant. The downwind area rank score was 16, and tests of probability reveal it to be significant at  $P < .0001$ . None of the rank sums for the upwind, right, and left quadrants were statistically significant.

In this analysis of the summer rainfall around St. Louis, I have considered the possible urban effect by using the prerin winds to define the probable placement of an urban-induced rain maximum. The fact that 9 out of 12 possible monthly (three summer rain months and four basic wind directions) patterns and three out of four summer patterns had downwind maximums is strongly suggestive of an urban influence on precipitation over and beyond the city. Rotating the rain with the winds reveals a 22 percent rain increase in the area downwind of St. Louis.

Statistical tests dealing with both the placement of the highest quadrant rainfall and the positions of rainfall by quantity showed that the downwind values were significantly different and higher than those of surrounding areas. These results for prerin wind directions and rainfall distribution around St. Louis strongly support the concept that major urban areas lead to increased summer rainfall.

STANLEY A. CHANGNON, JR.  
Illinois State Water Survey,  
Post Office Box 232,  
Urbana 61801

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10. The downwind versus right quadrant rainfall differences were significant at  $P < .046$ , and the downwind versus left quadrant differences were significant at  $P < .004$ .
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## Permeability of the Cell-to-Cell Membrane Channels in Mammalian Cell Junction

**Abstract.** *The channels in the junctions of various mammalian cell types—primary cultures and lines—were probed with a series of linear fluorescent amino acid and peptide molecules of different size and charge. Permeability is limited by probe size and electronegativity, these two factors apparently being related reciprocally. In respect to both factors, mammalian junctional channels are more restrictive than insect channels; hence the mammalian channels are narrower, more polar, or both. The channels of the various mammalian cell types differed slightly from each other; in some types the serum of the culture medium affected the channel permeability.*

Cells of organized tissues and cultures commonly form junctions through which hydrophilic molecules are transmitted from cell to cell (1-3). The elements in this junctional transmission are specialized, leakproof membrane channels that link one cell interior to the other (1, 3, 4). They are the largest biomembrane channels known. A study of an insect cell junction, that of *Chironomus* salivary gland, showed that they transmit probe molecules—linear amino acids and peptides—up to 1800 daltons (5). The limit for transmission in mammalian cell junction—which is structurally quite different (6)—is not known. We show here that the limit is lower.

We studied the junctions of the following cultured cell types: primary rat liver (i) epithelioid and (ii) fibroblastic (7), (iii) calf lens epithelioid, (iv) rat liver epithelioid line RL (8), (v) rat fibroblast line B (9), (vi) human mammary fibroblasts (HUMF) (8), mouse fibroblast lines (vii) 3T3-42 and (viii) 3T3-BALB/c, and (ix) hamster fibroblast line BHK. For a comparison with cultured insect cells we used the fibroblast lines (x) AC-20 from the homopteran leafhopper *Agallia constricta* and (xi) ATC-10 from the dipteran *Aedes aegypti* (10, 11).

Junctional permeability was tested with the same series of fluorescent, linear probes (Table 1) used before on insect junction (5), but with the series aug-

mented in the range 450 to 850 daltons. The molecules are of roughly constant abaxial dimension (14 to 16 Å, "width") and constitute a series of gradually increasing axial length. They are not degraded in cytoplasm (5), and their rates of loss are much less than their rates of transjunctional flux, in both the mammalian and insect cells (12). The 6-carboxyfluorescein supplements the series. This compound is more hydrophilic than fluorescein; like the labeled amino acids and peptides, it does not detectably permeate nonjunctional cell membrane from the outside (13). The probes were iontophoresed into the cells with the aid of a microelectrode, and their spread from the electrode through the injected cell and, transjunctionally, into the neighbors was continuously observed in a microscope dark field and photographed (14). To maximize sensitivity, we chose sparse cell groupings in which the injected cell was larger than the first-order neighbors, and made the fluorescence in the injected cell (roughly matched for all probes) much greater than the detection threshold. Permeability was scored positive when the tracer appeared in at least two neighbors, to make sure that it had not passed between incompletely divided daughter cells.

The outcome is summarized in Table 1. The feature that immediately stands out is the greater permselectivity of