Twins (human)

Two babies born to a mother at one birth. Knowledge about the biological bases of twinning, as well as sophistication in techniques for data collection, research design, and analysis, have increased substantially in recent years. Consequently, twin research has been incorporated into a growing number of behavioral science and medical science research programs.

Biology of twinning. There are two types of twins, monozygotic and dizygotic. Members of a twin pair are called co-twins. Controversy surrounding the definition of a twin arose with the advent of reproductive technologies enabling the simultaneous fertilization of eggs, with separate implantation. The 1996 cloning of Dolly the lamb in Scotland directed attention toward the promises and pitfalls of possible human cloning. The unique "twinlike" relationships that would result between parents and cloned children (who would be genetically identical to their parents) also challenge current conceptions of twinship. Monozygotic twins are clones (genetically identical individuals derived from a single fertilized egg), but parents and cloned children would not be twins for several reasons, such as their differing prenatal and postnatal environments. *See* REPRODUCTIVE TECHNOLOGY.

Monozygotic twins. The division of a single fertilized egg (or zygote) between 1 and 14 days postconception results in monozygotic twins. They share virtually all their genes and, with very rare exception due to unusual embryological events, are of the same sex.

A common assumption is that because monozygotic co-twins have a shared heredity, their behavioral or physical differences are fully explained by environmental factors. These environmental influences may occur during the prenatal, perinatal, or postnatal periods. However, monozygotic twins are never exactly alike in any measured trait, and may even differ for genetic reasons. For example, the random "shutting down" of one X chromosome in every female cell soon after conception (a process called lyonization) can cause monozygotic female twins to differ in X-linked traits, such as color blindness. *See* COLOR VISION; SEX-LINKED INHERITANCE.

Sometimes chromosomes fail to separate after fertilization, causing some cells to contain the normal chromosome number (46) and others to contain an abnormal number. This process, called mosaicism, results in monozygotic co-twins who differ in chromosomal constitution. These unusual cellular processes explain the presence of monozygotic pairs in which one co-twin is normal while the other shows a genetic anomaly reflecting a mixture of normal and abnormal cells. A rare case of monozygotic triplets including two healthy males and a co-triplet with Turner's syndrome (loss of one X chromosome) has been reported. *See* MOSAICISM.

There are several other intriguing variations of monozygotic twinning. Splitting of the zygote after day 7 or 8 may lead to mirror-image reversal in certain traits, such as handedness or direction of hair whorl. The timing of zygotic division has also been associated with placentation. Monozygotic twins resulting from earlier zygotic division have separate placentae and fetal membranes (chorion and amnion), while monozygotic twins resulting from later zygotic division share some or all of these structures. Compared with two-chorion monozygotic twins, monozygotic twins sharing a chorion are more likely to be born prematurely, to differ in birth weight, and to die early (in extreme cases). Associations between mirror-image reversal and placentation are expected, but relationships among these events do not appear to be straightforward. Should the zygote divide after 14 days, the twins may fail to separate completely. This process, known as conjoined twinning, occurs in approximately 1 monozygotic twin birth out of 200. The many varieties of conjoined twins differ as to the nature and extent of their shared anatomy. Approximately 70% of such twins are female. There do not appear to be any predisposing factors to conjoined twinning.

Dizygotic twins. Dizygotic twins result when two different eggs undergo fertilization by two different spermatozoa, not necessarily at the same time. Dizygotic twins share, on average, 50% of their genes, by descent, so that the genetic relationship between dizygotic co-twins is exactly the same as that of ordinary brothers or sisters. Theoretically, dizygotic twins may share between 0 and 100% of their genetic makeup, but most are close to the 50% average. Some dizygotic co-twins share higher or lower proportions of genes for certain traits, so they may be more or less similar in those traits. Dizygotic twins may be of the same or opposite sex, outcomes that occur with approximately equal frequency.

There are some unusual variations of dizygotic twinning. There is the possibility of polar body twinning, whereby divisions of the ovum prior to fertilization by separate spermatozoa could result in twins whose genetic relatedness falls between that of monozygotic and dizygotic twins, or between dizygotic twins and unrelated individuals. Blood chimerism, another variation, refers to the presence of more than one distinct red blood cell population, derived from two zygotes, and has been explained by connections between two placentae. In humans, chimerism can occur in dizygotic twins. New techniques estimate that chimerism occurs in 8% of dizygotic twins and 21% of dizygotic triplets. Superfecundation is the conception of dizygotic twins following separate fertilizations, usually within several days, in which case each co-twin could have a different father. Superfecundation may cause significant developmental discrepancies between co-twins due to their differing paternal heredity. Superfetation, which refers to multiple conceptions occurring several weeks or even one month apart, may be evidenced by delivery of full-term infants separated by weeks or months and by the birth or abortion of twin infants displaying differential developmental status. *See* FERTILIZATION; OOGENESIS.

Epidemiology. According to conventional twinning rates, monozygotic twins represent approximately one-third of twins born in Caucasian populations and occur at a rate of 3–4 per 1000 births. The biological events responsible for monozygotic twinning are not well understood. It is generally agreed that monozygotic twinning occurs randomly and not as a genetically transmitted tendency. Some recent evidence from Sweden suggests an increased tendency for mothers who are monozygotic twins to bear same-sex twins themselves; further work will be needed to resolve this question.

Dizygotic twinning represents approximately two-thirds of twins born in Caucasian populations. The dizygotic twinning rate is lowest among Asian populations (2 per 1000 births), intermediate among Caucasian populations (8 per 1000 births), and highest among African populations (50 per 1000 births in parts of Nigeria). The natural twinning rate increases with maternal age, up to between 35 and 39 years, and then declines. A possible causal mechanism is the increased ovarian activity that continues until a woman reaches her late thirties. Elevated levels of follicle-stimulating hormone (FSH) may increase the probability of dizygotic twinning among some women. Dizygotic twinning has also been linked to increased parity, or the number of children to which a woman has previously given birth. However, parity is associated with older maternal age, which is more closely linked to dizygotic twinning. Mothers of dizygotic twins are also significantly taller and heavier, on average, than mothers of monozygotic twins and singletons. *See* OVARY.

Dizygotic twinning appears to be genetically influenced, although the pattern of transmission within families is unclear. One study found that parents of both monozygotic and dizygotic twins are twice as often left-handed as their own same-sex singleton siblings, but the specific mechanism linking twinning and left-handedness is unknown. Recent work in New Zealand uncovered a gene in sheep associated with dizygotic twins and triplets that could assist the understanding of the genetics of human multiple birth.

A higher frequency of dizygotic twinning among lower social classes than among higher social classes has been reported. This relationship may simply reflect the larger size of the lower-class families. A relationship may exist between dizygotic twinning and dietary factors, but it could be quite complex: A reduction in the dizygotic twinning rate has been associated with reduced nutritional supply in some European countries during World War II, although there have been mixed trends regarding nutrition and overall twinning rates. Some researchers have implicated the yam (a food with estrogenlike substances) in the extremely high dizygotic twinning rate among the Yoruba of western Nigeria, although this is not definitive. Higher and more constant food supplies may explain the increased dizygotic twinning rate in southwest Finland's archipelago of Aland and Aboland, relative to the mainland.

Increased coital rate, following extended periods of sexual abstinence, may also increase the dizygotic twinning rate. The rate of dizygotic twinning is elevated in extramarital conceptions, during the first year of marriage, and in couples immediately after soldiers return from war. It may be that, in some women, coitus stimulates ovulation, and frequent ovulation leads to superfecundation. Increased dizygotic twinning has also been associated with an increased number of marriages.

The United States' twinning rate rose 65% between 1980 and 2002 and 38% between 1990 and 2002. It went up 3% between 2001 and 2002, to 31 twin births per 1,000 births. These increases were mainly due to advances in fertility treatments (for example, in vitro fertilization and ovulation induction), but were also due to delays in child-bearing. The increase mostly involved dizygotic twinning, in which multiple ovulation and maternal age are key factors. In vitro fertilization has, to a smaller extent, increased monozygotic twinning rates, possibly due to laboratory manipulation of the early embryo. However, in 2002 the birth rate of triplets and other higher order multiples dropped slightly to 184 per 100,000 births. This represented the third such decrease in the previous four years, following a 400% increase between 1980 and 1998.

The birth rate of preterm babies, defined as infants born after less than 37 weeks' gestation, increased 12.1% in 2002, and increased by 15% since 1990. This change partly reflects the increase in the rate of multiple births, which usually occur early.

Determination of zygosity. The accurate assignment of twin pairs as monozygotic or dizygotic is crucial for avoiding misleading estimates of genetic influence upon traits of interest. Co-twin comparison of selected DNA markers (six to eight) is the most accurate (greater than 99%) scientific method for classifying twins as monozygotic or dizygotic. Except for monozygotic twins, it would be extremely rare for family members to show matching DNA patterns. Cells for analysis can be obtained easily by gently scratching the inner cheek to obtain a buccal smear. Several laboratories are equipped to receive samples by mail and to provide twin-typing results in approximately 2 weeks. Blood-typing is another objective method for classifying twin pairs. Blood-group differences identify dizygotic co-twins with complete certainty. However, an absence of blood group differences can indicate only the probability that the twins are monozygotic, because in rare cases dizygotic co-twins may share all measured blood groups because of shared parentage. Analyses of anthropometric characteristics, such as fingerprint ridgecount (dermatoglyphic analysis), ponderal index, and cephalic index, improve the accuracy of twin classification when used in conjunction with blood-typing. *See* BLOOD GROUPS; DEOXYRIBONUCLEIC ACID (DNA); FINGERPRINT.

Other methods for diagnosing zygosity include physical-resemblance questionnaires and examination of placentae and placental membranes (**Fig. 1**). The more accurate of these methods is the physical resemblance questionnaire, which shows 93–99% agreement with the results from blood typing. Twins' and parents' judgments of twin type are often questionable.

Twin studies. Francis Galton reasoned late in the nineteenth century that comparative analyses of the behavioral and physical resemblance of the two types of twins would provide useful insights into the relative contributions of heredity and environment. (Although the biology of twinning was not established during Galton's time, he correctly recognized that there were two types of twins.) In particular, he suggested that greater resemblance within twin pairs derived from a single ovum (monozygotic twins), relative to twin pairs derived from separate ova (dizygotic twins), would demonstrate a contribution from genetic factors. This monozygotic–dizygotic twin comparison, called the classic twin method, assumes that differences between one-egg twins are generally due to environmental effects, given their shared heredity, and that differences between two-egg twins are due to genetic and environmental differences between them. The twin method additionally assumes that trait-relevant environmental influences are equal for both types of twins (equal environments assumption). Galton obtained detailed analyses of the life histories of twins, some of whom displayed early physical and behavioral similarities, and some an absence of early resemblance. He concluded that nature made a major contribution to many domains of human development.

Challenges to the classic twin method have been raised, despite its successful implementation in numerous behavioral science and medical science fields. Social biases refer to biases in treatment of monozygotic and dizygotic twins; for example, some critics of twin studies assert that there might be more similar treatment of monozygotic than dizygotic twins that may amplify behavioral similarity between the former and reinforce behavioral dissimilarity between the latter. Empirical testing has, however, demonstrated that similarity of treatment is unrelated to similarity of behavior among monozygotic twins. Primary biases refer to unique prenatal influences, such as shared intrauterine circulation, which tend to reduce resemblance between monozygotic co-twins. These effects do not, however, appear to seriously affect results from twin data based upon large samples. Recruitment biases refer to differences in representation among the types of twins who volunteer for research: approximately two-thirds of adult same-sex twin volunteers are female, and two-thirds are monozygotic. Twin studies carried out in Norway and the United States (Minnesota), however, were unable to demonstrate differences in resemblance for intelligence quotient (IQ), personality traits and interests, or demographic variables between solicited pairs in which both co-twins completed questionnaires by mail, as compared with pairs in which one twin participated. Twins who volunteer for research without solicitation, however, may not be representative of the general twin population. Another concern is that some twin studies are based on small samples, limiting the power of their conclusions.

Intraclass correlation. The intraclass correlation (r_i) is the preferred measure of resemblance between co-twins. It is based on a ratio of the variation between groups to the total variation (group refers to a twin pair) and is given by the equation below, where

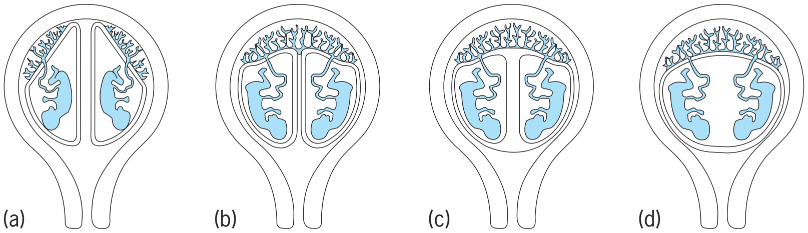

Fig. 1. Arrangements of placentae and placental membranes for monozygotic and dizygotic twin pairs in the uterus. (*a*) Monozygotic or dizygotic twins with separate amnions, chorions, and placentae. (*b*) Monozygotic or dizygotic twins with separate amnions and chorions and fused placentae. (*c*) Monozygotic twins with separate amnions and single chorion and placenta. (*d*) Monozygotic twins with single amnion, chorion, and placenta. Analysis of these arrangements can aid in diagnosing zygosity. (*After C. E. L. Potter*, *Fundamentals of Human Reproduction*, *McGraw-Hill*, *1948*)

$$r_i = \frac{S^2b - S^2w}{S^2b + (n-1)S^2w}$$

S^2b = variance between families, S^2w = variance within families, and n = average number of individuals per family.

Heritability expresses the proportion of population variation associated with genetic differences in a given characteristic. It is a function of the particular population under study, the time of measurement, and the measuring instrument, and is therefore not a constant figure but one that reflects changes in these factors. Subtracting the dizygotic intraclass correlation from the monozygotic intraclass correlation estimates half the genetic effect because dizygotic twins share half as many genes, on average, as do monozygotic twins. Doubling the resulting value yields an estimate of the genetic effect. This procedure works only if the genetic effects are additive; that is, the effects of the genes are constant, regardless of the genes with which they are paired. However, sometimes genes behave interactively or nonadditively—that is, the effects of the genes change depending upon the genes with which they are paired. This may increase the monozygotic twin intraclass correlation and heritability estimates because monozygotic twins always share gene combinations while dizygotic twins do not.

Methods for analyzing twin data have moved beyond analyses of correlations to more complex model-fitting approaches. Researchers develop a model specifying genetic and environmental relatedness and apply it to data from twins and other relatives. They can then estimate genetic and environmental contributions to the trait, based on how well the model fits the data.

Twin and adoption studies. Twins reared apart offer informative tests of genetic and environmental influences on behavior. If monozygotic twins are separated early in infancy and raised in separate homes selected at random, their degree of similarity (intraclass correlation) provides a direct estimate of genetic effects. The study of dizygotic twins reared apart enables additional tests of the possibility of genetic interactions. Another approach is to study

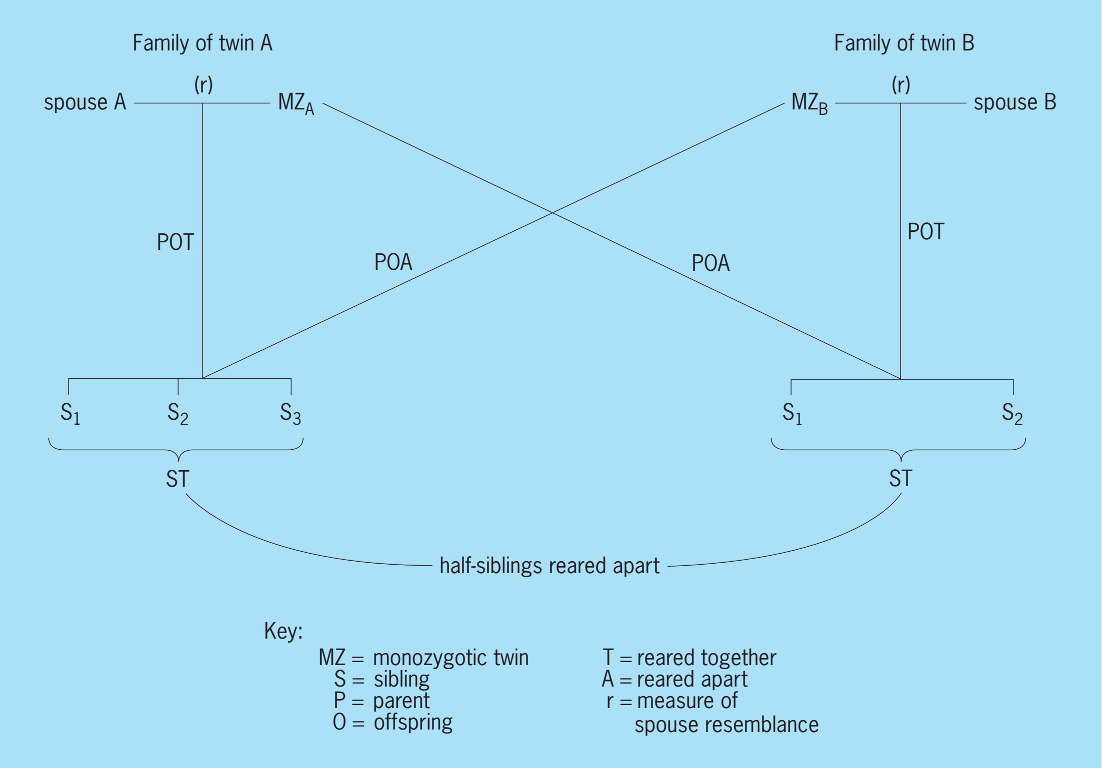

Fig. 2. Twin-family design. (*Adapted from S. Scarr-Salapatek, in K. W. Schaie, ed., Developmental Human Behavior Genetics*)

the adopted siblings of twins that are reared apart, a procedure that enables comparison between individuals who share environments but not genes; this is the reverse of studying separated monozygotic twins except that each twin and his/her adopted sibling are not the same age. This can be overcome by studying "virtual twins," genetically unrelated children of the same age raised together from infancy. They may consist of two adopted children or one adopted child and one near-in-age biological child in the family. They share their environments but not their genes so they truly are the reverse arrangement of monozygotic twins reared apart. One laboratory in the United States (at California State University, Fullerton) is studying these "twinlike" sibships to determine the extent to which common environments affect behavioral similarities.

Comparison of resemblance between monozygotic and dizygotic twins reared together and those reared apart also provides information on the influence of common rearing on behavioral and physical similarity. The level of personality resemblance is similar for monozygotic twins reared apart and reared together (approximately 50% of the variance is associated with genetic factors), demonstrating that common environmental influences do not contribute importantly to personality similarity. Resemblance in the body mass index (which is the weight in kilograms divided by the height, in squared meters) is only slightly lower for monozygotic twins reared apart than for monozygotic twins reared together. These analyses suggest that both genetic factors and environmental factors unique to the individual are important influences on these characteristics.

Twin gender studies. There has been recent interest in gender-related behaviors of opposite-sex twins. This is due to findings in the nonhuman literature showing that female mice and gerbils demonstrate masculine behaviors if situated next to a male fetus in utero. It is suggested that prenatal exposure to male hormones may be responsible for these results. Human twin research has produced mixed findings in this regard. It is also important to consider that female co-twins' behaviors could reflect the psychological and social effects of being raised with a same-age male sibling. However, the finding that females with male co-twins show the same frequency of otoacoustic emissions ("hums" discharged in the inner ear to raise the volume of weak sounds) as their brothers is difficult to explain without reference to opposite-sex twins' prenatal biological circumstances.

Twin-family studies. The offspring of monozygotic twins share unique relationships with one another. The children of monozygotic twins are genetically equivalent to half-siblings, because they each have a parent that is genetically identical (either a twin mother or a twin father). Similarly, monozygotic twins share the same genetic relationship with the co-twin's children (nieces and nephews) as they do with their own children. This family constellation thus enables comparisons between co-twins, marital partners, twins and children, twins with nieces and nephews, full siblings, and half-siblings. Studies of mental abilities and physical characteristics that use the twin-family method are available. Studying social relationships among members of these unique families is also potentially informative with respect to genetic and social influences (**Fig. 2**).

Twins as couples. This approach to twin research compares monozygotic and dizygotic twins' social relationships with reference to their joint behaviors, such as cooperation and competition. The focus of interest is the behavior of the pair (for example, their level of cooperation or competition), not their separate behaviors as in classic twin studies. Research using this design has generally revealed greater cooperation between monozygotic twins than dizygotic twins in experimental games and tasks. These tendencies mirror the increased social closeness that monozygotic twins typically express toward one another and the increased grief they experience following the loss of their co-twin, relative to dizygotic twins. There is, however, overlap among twin types—some dizygotic twins are especially close to one another, while some monozygotic twins are less so.

QTL studies. Quantitative traits, such as height, intelligence, and certain aspects of personality, are influenced by many genes working together. People vary continuously on such traits, ranging from short to tall; average to highly intelligent; and outgoing to shy. Quantitative trait loci (QTLs) are the many genes that affect these traits, although they each differ in their effect. Identifying genes with the greatest influence on traits of interest can tell us about individual differences in the development of that trait. Now that the Human Genome Project has mapped the full complement of 20,000 to 25,000 genes, behavioral scientists and molecular geneticists have increased their efforts at finding QTLs for mental retardation, aggression, and psychopathology. It has how been shown that individuals with the apolipoprotein e4 gene are at increased risk for late-onset Alzheimer's disease. Twin studies will play important roles in the quest to find QTLs. The most valuable information may come from studying monozygotic twins who differ in their expression of disease and behavior. In the case of twins differing in schizophrenia, it may be that one twin was exposed to specific environmental factors that activate relevant genes or QTLs, while the other twin was not. Furthermore, even monozygotic twins are not genetically identical, as indicated above. Unraveling the link between these genetic differences and behavior will be an informative, albeit challenging task. *See* HUMAN GENOME PROJECT.

Twin registries. Twin studies depend on large samples in order to draw valid conclusions about behavior and development. Statistically significant findings are those occurring by chance fewer than one time in twenty. Statistical significance is associated with sample size and the size of the correlation. A .25 correlation would be statistically significant with 45 twin pairs, while a .50 correlation would be statistically significant with 12 twin pairs. Large twin samples are also required for estimating heritability, especially if the heritability of a given trait is low in the population. However, a large number of subjects does not ensure valid findings because the quality of the information may be difficult to determine. It has also been shown that heritability may be calculated more accurately and efficiently using twins reared apart rather than twins reared together. For example, 400–500 monozygotic and 400–500 dizygotic twin pairs reared together allow heritability to be estimated with the same degree of confidence as 50 monozygotic twin pairs reared apart. This is because heritability is estimated directly from the reared apart twins and indirectly from the reared together twins.

Modern twin research has come a long way in recruiting twins since Francis Galton did his famous study of nature and nurture in 1875. Galton gathered twins by sending notices to twins and relatives of twins. Respondents were asked to provide the names and addresses of other twins that they knew, thus enlarging his potential participant pool. Galton received 80 replies, although he did not indicate what percentage of his initial inquiries this number represented. Thirty-five twin pairs, presumably monozygotic, showed "close similarity" based on written responses to his questions. Galton clearly showed that twin studies could illuminate genetic and environmental influences on human behavior. However, findings from small samples gathered unsystematically urge cautious interpretation of the results. Fortunately, there is now a growing number of population-based twin registries.

The oldest twin registry in the world is the Danish Twin Registry, established in 1954. It now includes over 65,000 twin pairs, comprising 127 birth cohorts. A recent review summarized characteristics for 16 national twin registries, located in the United States, Australia, Belgium, Canada, China, Denmark, Finland, Germany, Italy, Japan, Korea, the Netherlands, Norway, Sri Lanka, Sweden, and the United Kingdom. There are also statewide twin rosters such as the Minnesota Twin Registry and the Wisconsin Twin Panel, and commercial registries such as the United Kingdom's *Gemini Genomics*, dedicated to finding disease-related genes. Investigators interested in specific conditions can try to identify specific subjects within these sources. There are, however, registries that target twins with particular conditions such as HIV exposure and chronic fatigue syndrome. Some registries include specific subgroups of twins, such as World War II veterans, Vietnam veterans, and elderly citizens. Aside from offering researchers access to large and diverse twin samples, the registries facilitate international collaboration. Studies of rare medical conditions can proceed by pooling cases across sites. Attempts at cross-cultural replication of findings can also be conducted.

Value of twin studies. Studies of monozygotic and dizygotic twins provide valuable information on genetic and environmental influences on human behavioral and physical development. Studies of twins, therefore, offer many opportunities for examining novel hypotheses and generating new explanations of observations. For example, twin studies have been used to assess predictions generated by human evolutionary biology and theories of economics. New model-fitting approaches to data analysis enable testing for the presence of specific genetic and environmental effects. Refinement of twin samples and the study of more well-defined characteristics is another important trend. In recent years, twin research has begun to address the origins of less frequently studied behaviors such as humor, love styles, athleticism, and religiosity. Continued extension and elaboration of the twin design, replication of research, and continued testing of assumptions will help researchers to improve twin methods and respond to criticism. *See* BEHAVIOR GENETICS; HUMAN GENETICS.

Nancy L. Segal

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