Breeding Technologies to Increase Crop Production in a Changing World

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To feed the several billion people living on this planet, the production of high-quality food must increase with reduced inputs, but this accomplishment will be particularly challenging in the face of global environmental change. Plant breeders need to focus on traits with the greatest potential to increase yield. Hence, new technologies must be developed to accelerate breeding through improving genotyping and phenotyping methods and by increasing the available genetic diversity in breeding germplasm. The most gain will come from delivering these technologies in developing countries, but the technologies will have to be economically accessible and readily disseminated. Crop improvement through breeding brings immense value relative to investment and offers an effective approach to improving food security.

Although more food is needed for the rapidly growing human population, food quality also needs to be improved, particularly for increased nutrient content. In addition, agricultural inputs must be reduced, especially those of nitrogenous fertilizers, if we are to reduce environmental degradation caused by emissions of CO₂ and nitrogenous compounds from agricultural processes. Furthermore, there are now concerns about our ability to increase or even sustain crop yield and quality in the face of dynamic environmental and biotic threats that will be particularly challenging in the face of rapid global environmental change. The current di-

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more so given the additional pressures created by global environmental changes.

Global Environmental Change Alters Breeding Targets

Certain aspects of global environmental change are beneficial to agriculture. Rising CO₂ acts as a fertilizer for C₃ crops and is estimated to account for approximately 0.3% of the observed 1% rise in global wheat production (4), although this benefit is likely to diminish, because rising temperatures will increase photorespiration and nighttime respiration. A benefit of rising temperatures is the alleviation of low-temperature inhibition of growth, which is a widespread limitation at higher latitudes and altitudes. Offsetting these benefits, however, are obvious deleterious changes, such as an increased frequency of damaging high-temperature events, new pest and disease pressures, and altered patterns of drought. Negative effects of other pollutants, notably ozone, will also reduce benefits to plant growth from rising CO₂ and temperature.

Particularly challenging for society will be changes in weather patterns that will require alterations in farming practices and infrastructure; for example, water storage and transport networks. Because one-third of the world’s food is produced on irrigated land (5, 6), the likely impacts on global food production are many. Along with agronomic- and management-based approaches to improving food production, improvements in a crop’s ability to maintain yields with lower water supply and quality will be critical. Put simply, we need to increase the tolerance of crops to drought and salinity.

In the context of global environmental change, the efficiency of nitrogen use has also emerged as a key target. Human activity has already more than doubled the amount of atmospheric N₂ fixed
anually, which has led to environmental impacts, such as increased water pollution, and the emission of greenhouse gases, such as nitrous oxide. Nitrogen inputs are increasingly being managed by legislation that limits fertilizer use in agriculture. Furthermore, rising energy costs means that fertilizers are now commonly the highest input cost for farmers. New crop varieties will need to be more efficient in their use of reduced nitrogen than current varieties are (7). Therefore, it is important that breeding programs develop strategies to select for yield and quality with lower nitrogen inputs.

Current Approaches to Crop Improvement

Arguably, increased yield in conditions of abiotic stresses, such as drought and salinity, could be best achieved by selecting for increased yield under optimal production conditions: Plants with higher yields in good conditions are more likely to have higher yields in stressed conditions (8). Such an approach will also increase yield in high-yield environments. However, it is becoming increasingly apparent that specific selection strategies are needed to enhance yield in low-yielding environments. Given that average global yields of wheat are less than 3 metric tons/ha (Fig. 1) and given there are many areas with yields as high as 10 metric tons/ha, the majority of land cropped to wheat delivers yields below 3 metric tons/ha. Therefore, by virtue of the much larger areas of low-yielding land globally, low-yielding environments offer the greatest opportunity for substantial increases in global food production. Increasing yield by 1 metric ton/ha in a low-yielding area delivers a much higher relative increase than does the same increase in high-yielding environments. This increase can be achieved by tackling major limitations on yield in poor environments (termed yield stability); for example, by protecting plants and yield from factors such as salinity and heat or drought periods. The local social benefits of supporting farmers on low-yielding lands would also be great.

It is often thought that concentration on yield stability may come at the expense of high yields in good years; however, yield penalties in more favorable conditions do not necessarily accompany drought tolerance (Fig. 2).

Yield stability is harder to select for than improved yield is, because selection in breeding programs requires many years and many sites for evaluation. However, there is evidence for a genetic basis for yield stability and, hence, an opportunity for gain (9). Transgenic approaches are also likely to improve yield stability (10). There are several clear examples where single genes have been able to substantially increase yield, notably to drive domestication (to control tiller number, branching, and seed number) and the green revolution (for dwarfing). Initial results suggest that a gene conferring increased drought tolerance may also have a widespread impact on yield (10).

This is not to say that efforts to maintain yield should be reduced. In particular, maintaining resistance to rapidly evolving pests and pathogens is an essential mainstay of breeding programs. Interactions between breeders, pathologists, and agronomists must be maintained to ensure that crops and cropping systems change coordinately. No-till farming, in which plowing of the soil is avoided, for example, has changed the spectrum of diseases and pests attacking crops, to the extent that a change in breeding targets was needed. The development of multiple cropping systems will also demand interactions between agronomists.
Box 1. New breeding technologies.

MAS uses a marker such as a specific phenotype, chromosomal banding, a particular DNA or RNA motif, or a chemical tag that associates with the desired trait. For example, a DNA marker closely linked to a disease resistance locus can be used to predict whether a plant is likely to be resistant to that disease.

- Gene pyramiding can usually only be accomplished by using MAS. For example, pyramiding is used to create durable disease resistances by selecting for two or more resistance genes against a pathogen. Multiple, partial, rust-resistance genes in wheat can be accumulated into elite varieties to provide strong and durable resistance. Single genes would give only weak resistance, and MAS offers the only effective method for accumulating multiple resistances (22).

- Marker-assisted recurrent selection (MARS) involves crossing in selected individuals at each cycle of crossing and selection. In this way, desirable alleles can be brought into the breeding scheme from many different sources. This technique has been applied to sunflower, soybean, and maize to bring desirable alleles at several target loci into single elite lines (27).

- Genome-wide or genomic selection also relies on MAS and is under evaluation for the feasibility of incorporating desirable alleles at many loci that have small genetic effect when used individually. In this approach, breeding values can be predicted for individual lines in a test population based on phenotyping and whole-genome marker screens. These values can then be applied to progeny in a breeding population based on marker data only, without the need for phenotypic evaluation. Modeling studies indicate that this method can lead to considerable increases in the rates of genetic gain by accelerating the breeding cycles (20). In the oil palm, for example, this approach could lead to the release of improved germplasm after only 6 years as compared with the current time of 19 years (28).

- Complex trait dissection uses high-throughput technologies to determine the phenotypic components of complex traits. For example, robotic greenhouse systems use nondestructive imaging to monitor growth rates, stem and leaf architecture, and root structure (for example, see www.lemnatec.com/). Similar systems can also be adapted for the detection of characteristics of chlorophyll fluorescence (which indicate aspects of plant responses to the environment) or fluorescent protein–labeled genotypes.

- The analysis of complex traits has recently been bolstered in statistical and modeling methods for the analysis of phenotypic data obtained from field and controlled environment studies. For example, in assessing drought tolerance in wheat and sorghum, modeling can be used to generate an "index of the climatic environment" to identify the stages of crop development where there is the strongest interaction between genotype and the environment and to identify aspects of the crop response that can be most readily enhanced by breeding and selection (29).

- Increasing genetic diversity requires an expansion of the germplasm base in breeding programs (22), but this is dependent on enhancing techniques for assessing the value of the program and using individual accessions from germplasm collections. Improvements in phenotyping and genotyping will help remove this limitation by facilitating the identification and characterization of key adaptive QTLs. For example, increased expression of a boron transporter in a barley landrace leads to high tolerance to soil boron in elite varieties when the high-expression allele is transferred. Screening for variation in expression levels for this gene in germplasm collections may identify new sources of tolerance (30).

- Introgression of novel alleles from landraces and wild relatives is often slow and tedious, but options are now being developed for accelerating introgression as we learn more about the recombinational behavior of plant genomes and develop new breeding methods.

- The wider deployment of GM approaches will be needed for the introduction of novel genes and alleles from diverse sources, and particularly for traits that are absent from plant genomes (for example, Bacillus thuringiensis toxin from soil bacteria) or where there is insufficient variation for practical utility (for example, vitamin A accumulation in rice endosperm).

- The constraints on regulatory and consumer acceptance of GM can be reduced by adopting alternative approaches for engineering plants. For example, consumer acceptance may be greater and regulatory approvals simpler for plants transformed with cis-genic vectors in which only host gene sequences are used in the transformation construct (www.cisgenics.com). Similarly, the creation of marker-free plants, where only the DNA that has a biological effect remains in the plant, has been used to develop plants without antibiotic-resistance genes, which has caused much controversy (31).

- Heterosis (hybrid vigor) for inbreeding species (that is, species that usually self-pollinate, such as rice and wheat) can offer 20% to over 50% yield increases, and, for example, a 68% increase in yield has been achieved in foxtail millet (32). Strategies for using heterosis more widely to increase yields in inbreeding crops center on finding ways of reducing the cost and increasing the efficiency of producing hybrid seed. These include identifying new sources of male sterility for hybrid creation (such as thermosensitive genic male sterility in rice (33)) and using GM approaches to engineer sterility and restore fertility (such as the InVigor Canola from Bayer CropScience). Another possible mechanism for producing hybrid seed involves the use of apomixis, where plants produce seed without the need for fertilization. This allows hybrid vigor to be fixed. Creating apomictic crop plants may also be possible as we learn more about the genes controlling this process.

- Direct targeting of key heterotic loci may also be achievable as we learn more about the molecular basis of hybrid vigor (for example, in maize) (34).

Limitations

Of course, none of this will happen without suitably trained staff in plant breeding and molecular biology, so substantial increases in the education of plant breeders are essential. Most countries are struggling to maintain strong breeding capabilities. A vital adjunct is the free communication of resources and capabilities from technology developers to technology users. Resource and capacity building within breeding programs is essential to develop novel approaches, particularly in developing countries. Furthermore, developing countries critically need support for the development of crops, where there has been little interest from the developed world and, consequently, little investment. In many cases, these "orphan crops," such as cassava and plantain, are of critical importance for food security.

For many of the new breeding technologies, access to equipment, reagents, and skilled personnel is critical. Whereas service providers deliver this support to breeding programs in some parts of the world, they are often too expensive for poorly resourced breeding programs, and the logistics of sending plant tissue samples for analysis in a timely fashion can be prohibitive. Some organizations are attempting to address this limitation by establishing support services for breeding programs in the developing world (www.generationcp.org/).
and breeders. However, it is clear that more is required than can be provided by traditional breeding approaches.

**Emerging Technologies for Crop Breeding**

The production and evaluation of genetically modified (GM) crops is an active area of research, but the access of growers to this technology in many countries is currently restricted primarily because of political and bioethical issues (Box 1). Nevertheless, GM technologies permit the generation of novel variation beyond that which is available in naturally occurring (or even deliberately mutated) populations. Classic applications of GM include the use of proteinaceous toxins to control insect pests and "golden rice," which is biofortified with vitamin A (11). Crucial to the future deployment of GM crops are the discovery and characterization not only of genes but of promoters that provide accurate and stable spatial and temporal control of the expression of the genes (12).

Development of cis-genic vectors and marker-free transgenic plants (Box 1) may help to ease some of the political concerns about GM technologies. Nevertheless, the widespread application of GM technologies will remain limited while regulatory demands impose high costs on releasing GM crops (Box 1). Although it is likely that most of the important contributions to crop improvement in the coming 5 to 10 years will continue to be from non-GM approaches, we consider that transgenic technologies will inevitably be deployed for most major crops in the future.

Methods of crop breeding have undergone major changes, and a range of technologies is improving the rate and success of crop improvement in some breeding programs, but these have yet to be widely adopted. Contributions are being made through new selection strategies that are informed by sophisticated genetics, the use of computers to track and manage field trials, and biometric methods for field-trial design and assessment of interactions between genotype, environment, and management (13).

Marker-assisted selection (MAS) techniques (Box 1) are free of the political issues that have plagued the application of GM technologies. MAS involves using variation at the DNA level to track and monitor specific regions of the genomes during crossing and selection (14). The greatest benefit of MAS occurs where the target traits are of low heritability, are recessive in nature, and involve difficult and costly phenotyping, and where pyramiding of traits is desired for results such as disease and pest resistance. In these cases, MAS is likely to be more reliable, more convenient, or cheaper than phenotype-based selection, and MAS currently provides the only viable method for gene pyramiding. Molecular markers are also important in analyzing the mode of inheritance of certain traits and assessing genetic diversity. In cases where desirable traits are closely linked and in repulsion, markers can be critical in selecting rare recombination events.

In many cases, MAS provides an important alternative to phenotypic selection. However, the success of markers depends on their reliability in predicting phenotype. Many key stresses associated with rapid environment changes, notably drought and salinity tolerance, are complex and highly variable. For these types of traits, it is necessary to dissect tolerance into component contributory traits and to identify genetic regions encoding the traits, rather than overall plant tolerance (6, 15, 16). However, this genetic approach requires high-throughput phenotyping (phenomics) (17) (Box 1). Phenomics also allows screening of populations for particular traits and will facilitate the introgression of novel variation from wild germplasm. Phenomics will enable tighter definition of the properties of molecular markers, allowing introgression of appropriate combinations of tolerance traits into commercial varieties for particular target environments.

The combination of reliable phenotyping and MAS has been particularly important in transferring desirable alleles by simple backcrossing into elite germplasm. Although MAS has been used to track multiple independent loci (18), conventional breeding schemes become quite complex as the number of target loci expands. To overcome the problems of dealing with multiple loci, in particular, multiple loci of small genetic effect, two relatively new methods involving MAS can be deployed: marker-assisted recurrent selection (MARS) and genome-wide or genomic selection (GWS) (19, 20) (Box 1). MARS involves crossing selected individuals at each selection cycle so that desirable alleles at the target loci are introduced one at a time or through the merging of multiple crossing and selection streams. A problem with this approach is that it is most effective for genes or quantitative trait loci (QTLs) of major effect. In contrast, GWS does not require prior information on marker trait associations and can be used to select for multiple loci of small genetic effect. In this approach, populations are extensively genotyped to give full genome coverage and phenotyped. Subsequently, these data allow the prediction of phenotypic performance of an individual on the basis of whole-genome marker surveys.

These new breeding and selection strategies rely on the availability of cheap and reliable marker systems. A serious limitation in marker application for some species has been the paucity of useful markers. However, the new sequencing platforms have allowed large-scale discovery of single-nucleotide polymorphisms (SNPs) for species where few markers were previously available. The new marker systems combined with the new marker-based selection and screening strategies provide a base for a revolution in crop breeding and genetics.

**Expanding the Germplasm Base for Plant Breeding**

The success of plant breeding over the past century has been associated with a narrowing of the available genetic diversity within elite germplasm, particularly for some species such as peanut and soybean. New sources of variation include landraces and wild relatives of crop species, and although exploiting wild relatives as a source of novel alleles is challenging, it has provided notable successes in crop improvement. A particularly important example of the introgression of genetic information from a relative was the use of the short arm of rye chromosome 1R in wheat. In the early 1990s, this wheat-rye translocation was used in 45% of 505 bread wheat cultivars in 17 countries (21). Increasingly easy gene discovery, improved enabling technologies for genetics and breeding, and a better understanding of the factors limiting practical exploitation of exotic germplasm promise to transform existing, and to accelerate the development of new, strategies for efficient and directed germplasm use (Box 1).

Most crop geneticists agree that enrichment of the cultivated gene pool will be necessary to meet the challenges that lie ahead. However, to fully capitalize on the extensive reservoir of favorable alleles within wild germplasm, many advances are still needed. These include increasing our understanding of the molecular basis for key traits, expanding the phenotyping and genotyping of germplasm collections, improving our molecular understanding of recombination in order to enhance rates of introgression of alien chromosome regions, and developing new breeding strategies that permit introgression of multiple traits (22). Recent progress has shown that each of these challenges is tractable and within reach if some of the basic problems limiting the application of new technologies can be tackled.

**Limitations in Applying the New Technologies**

Several issues are likely to limit the application of these new methods, particularly for breeding programs in the public sector (Box 1). Regulatory complexity and high costs have prevented the widespread delivery of GM technologies (Box 1). Over the coming decade or so, however, it seems inevitable that GM technologies will become much more widely used—it is probably a case of “when,” not “if.” A consequence emerging for crops that are now dominated by GM varieties (such as cotton, soybean, and maize) is that breeding programs are now based around GM varieties, and consequently, breeding programs in non-GM jurisdictions have limited access to current advances. The key limitations for traditional breeding include lack of resources, training, and capabilities for most of the world’s
crop improvement programs (23, 24) (Box 1). It is important, therefore, that we expand the scope of access to new marker platforms to provide efficient, cost-effective screening services to the breeders. Communication and mechanisms for delivery of material to breeders must be developed. There is an urgent need to expand the capacity of breeding programs to adopt new strategies. The clearly documented high rate of return on such investments in the past should be kept in mind (25).

The concerns about food security and the likely impact of environmental change on food production have injected a new urgency into accelerating the rates of genetic gain in breeding programs. Further technological developments are essential, and a major challenge will be to also ensure that the technological advances already achieved are effectively deployed.

References and Notes
23. The Global Partnership Initiative for Plant Breeding can be found at http://gpi.ornl.gov/.
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PERSPECTIVE

Smart Investments in Sustainable Food Production: Revisiting Mixed Crop-Livestock Systems

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Farmers in mixed crop-livestock systems produce about half of the world’s food. In small holdings around the world, livestock are reared mostly on grass, browse, and nonfood biomass from maize, millet, rice, and sorghum crops and in their turn supply manure and traction for future crops. Animals act as insurance against hard times, and supply farmers with a source of regular income from sales of milk, eggs, and other products. Thus, faced with population growth and climate change, small-holder farmers should be the first target for policies to intensify production by carefully managed inputs of fertilizer, water, and feed to minimize waste and environmental impact, supported by improved access to markets, new varieties, and technologies.

“B”usiness as usual” investments in agriculture, although necessary (1, 2), are unlikely to deliver sustainable solutions as the world rapidly changes (3, 4). At the recent GS summit in Italy, the leaders of the world’s wealthiest countries promised to invest U.S.$20 billion to improve global food security. Most of that money is likely to flow to the developing world, where over the next few decades agricultural systems, already facing a variety of stresses, will be expected to accommodate a massive population surge. Even an investment of this magnitude could fail to generate food security if its deployment is not well planned and based on sound science.

The usual culprits, such as inefficient aid delivery, government corruption, and political unrest, are a barrier to progress but are not the most important problem. Rather, it involves a fundamental failure to appreciate the range of different agricultural systems that are expected to feed our planet in the coming decades and their policy needs. The diverse pressures that are acting on agricultural systems in various parts of the world include population increase, rising incomes and urbanization, a rapidly rising demand for animal products in many developing countries, and a fierce competition for land and water (3, 5, 6), all of which will have profound effects on food security (7). Croppers and livestock keepers the world over have steadily accumulated local experience and knowledge that will help them to adapt in the future, but the rapid rates of change seen in many agricultural systems in developing countries may simply outstrip their capacity. Yet, recent scientific assessments (1, 2, 7–10) and the technical and policy recommendations that flow from them have not fully captured the complex biological, social, and economic dynamics of the variety of chal-

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A Kinder, Gentler Jeremy Rifkin Endorses Biotech, or Does He?

For years, activist Jeremy Rifkin was the bête noire of biotechnology. Beginning in 1983, he filed several lawsuits to block field trials of genetically modified (GM) organisms and grabbed headlines around the world. Rifkin, an economist who runs the nonprofit Foundation on Economic Trends in Washington, D.C., said such actions were necessary to force an insulated research world to confront pressing ethical questions. To many in the scientific community, however, Rifkin was simply fanning irrational fears about biotechnology. A headline of a 1989 *Time* magazine profile called him “The Most Hated Man in Science” and captured the prevailing sentiment.

After a decade and a half of protests and campaigns to ban GM crops, Rifkin largely moved on to other topics, such as commerce, European politics, and hydrogen fuel. But now Rifkin, 61, is jumping back into agricultural biotech—this time, as a promoter. “This is an amazing twist for Jeremy Rifkin,” says Susan McCouch, a rice geneticist at Cornell University. “I’ve never seen the man come out in favor of anything.” But, like many others, she doubts his support will make much difference, as he is endorsing a biotech approach, known as marker-assisted selection (MAS), that is already well accepted.

In a white paper posted to his organization’s Web site* this week, Rifkin says MAS offers all the advantages of new genomic science without what he calls the great risks to human health and the environment posed by GM crops. Instead of transferring genes from one species to another, MAS simply speeds and improves traditional plant breeding. Researchers search through maps of a plant’s genome for sequence markers that fit his agenda. “To say that marker-assisted breeding or that MAS has somehow rendered it obsolete is simply wrong,” says Alan McHughen of the University of California, Riverside. “He still twists information to fit his agenda.” Rifkin does indeed argue that GM crops should be phased out. He claims few crops have been improved by transgenic modification—“it’s primitive science”—and, to make matters worse, contamination of wild relatives by transgenes may complicate the process of MAS, he warns.

As Rifkin describes it, his conversion was gradual. After following MAS for some time, he says he realized last year that it had eclipsed transgenic technology in its potential. MAS certainly has provided an enormous boost to breeders, and the pace has accelerated as ever more DNA is sequenced and as genetic screens have become cheaper and faster. Although scientists and companies share Rifkin’s enthusiasm for MAS and predict it will become even more powerful, they disagree that transgenic technology has failed or that MAS has somehow rendered it obsolete. “To say that marker-assisted breeding will replace biotech is simply wrong,” says Roger Beachy, who directs the Donald Danforth Plant Science Center in St. Louis, Missouri. That’s because of the enormous task facing plant breeders, says Mike Gale, an emeritus cereal geneticist at the John Innes Centre in Norwich, U.K.: “If we are going to produce enough food to feed the world, we need every tool in the toolbox.”

McCouch agrees that gene splicing remains a crude approach—like adjusting an intricate watch with a sledgehammer. Yet, she and others say, it is the only way forward in some cases—for instance, if a gene for a particular trait can’t be found in a crop or its wild relatives. The classic example is Bt, a toxin from a soil bacterium that was added to corn to provide broad and powerful protection against lepidopteran insects. Now companies are working to add genes for omega-3 fatty acids into soybean, to make the oil more healthful. “Those genes don’t exist in soybeans at all,” says David Fischoff, head of technology strategy and development at the Monsanto Co. in St. Louis, Missouri.

Nor is transgenic technology inherently risky, scientists say. “It is the gene and the management of the crop that make the difference and not the technology used to develop them,” says Les Firbank of the Centre for Ecology and Hydrology in Lancaster, U.K.

Rifkin’s concerns aren’t just biological. He couples his endorsement of MAS with a few caveats about policy, as well. He wants to be sure the technology is used in a way that meets its broader goals of sustainable agriculture and open-source technology—in other words, no patents. “We’ve seen too much how the patent system restricts the
cooperative nature in science,” he says. Charles Benbrook, a scientist with the Organic Center in Enterprise, Oregon, agrees that tight constraints on intellectual property are a concern, as ever more technology and markers are locked up in company labs. “I worry that marker-assisted breeding is not going to be able to deliver on its potential.”

Although Rifkin stops short of calling for an overhaul of patent law, he predicts that genetic technology and genomic information will eventually make it so easy and cheap to produce germ plasm that companies will have to make profits by selling agroecological consulting to farmers. Rifkin says he plans to start actively hawking his message on the lecture circuit and in his advice to business leaders and governments. “This is what I’m going to hammer away on: MAS should be phased in on the condition of an agroecological approach and open source.”

Rifkin’s pleas aside, Monsanto and other agribusiness companies contacted by Science don’t plan to drop their GM research or stop seeking patents. And several in the scientific community say they don’t need Rifkin’s help promoting a field that’s already flourishing. “Having the endorsement of Jeremy Rifkin means nothing,” says Martina Newell-McGloughlin, director of the University of California’s Biotechnology Research and Education Program in Davis. She and others doubt that any conversation with Rifkin would be productive. “Let’s just ignore the man,” says Gale. “Let’s get on with the job we have, which is to feed the world.” But whether or not Rifkin succeeds in opening the conversation he desires, he no doubt will keep talking.

—ERIK STOKSTAD

PALEONTOLOGY

Fossil Embryos Hint at Early Start for Complex Development

Evidence of the earliest animals on Earth dates back about 700 million years. But the arrival time of more complex animals—those with mirror symmetry and digestive tracts, known as bilaterians—has remained a mystery.

Now, on page 1644, an international team of paleontologists says it has isolated hundreds of fossil embryos that resemble those of modern bilaterians such as annelids and mollusks. If they check out, it could mean that a wide array of complex animals existed tens of millions of years before the “Cambrian explosion”—the time when paleontologists think hard-bodied animals proliferated as their ecosystems took shape. Precambrian animals have been notoriously difficult to find and study because their fragile bodies likely did not fossilize well. “I’m delighted to see a paper like this because it suggests there’s more to look for out there,” says Rudolf Raff of Indiana University in Bloomington.

Two years ago, Jun-Yuan Chen of Nanjing University in China and colleagues described fossils with bilaterian features in Science (9 July 2004, p. 218). The finds, uncovered in 580-million- to 600-million-year-old rocks in the Doushantuo deposit in China’s Guizhou Province, drew fire from paleontologists who suggested that the small, almost featureless “fossils” were actually layers of minerals. The latest discovery comes from the same rocks, but this time Chen’s team has unearthed what appear to be fossil embryos bearing hallmarks of bilaterian embryos. If they are bilaterians, says Jon Mallatt of Washington State University in Pullman, it would mean these complex animals existed 40 million years earlier than current evidence suggests.

The researchers say the fossil embryos sport so-called polar lobes, asymmetrical bulges that allow bilaterian embryos to form different tissues in adults. The fossil embryos appear to be in different stages of development: Some have three lobes and some five; some have lobes of equal shape, and some are more lopsided. Chen and colleagues isolated the fossils by dissolving away surrounding rock with acid, then examined them under a scanning electron microscope. The researchers say the relative volumes of the spherical lobes are too regular for the fossils to be unrelated embryos or inorganic lumps of rock stuck together.

Some scientists are reluctant to give the fossils their unequivocal endorsement. Nicholas Holland, an invertebrate zoologist at the University of California, San Diego, notes that the specimens show a few “slightly bothersome” differences from other bilaterian embryos. “A lot of critters that make polar lobes have reasonably small eggs, around 200 microns,” says Holland, whereas many of the Doushantuo embryos are as much as five times bigger.

Douglas Erwin of the National Museum of Natural History in Washington, D.C., cautions that the embryos’ small size makes it hard for scientists to tell organic structures from mineral deposits and other preservation-related artifacts. “If you have a dinosaur bone, it’s easy to tell what’s bone or what’s not,” he says. “The closer you look, the harder it is to tell what the original bone structure is.” The fossils also lack some characteristics of known annelids and mollusks, Erwin says, although the embryos could represent an extinct lineage of bilaterian.

Just knowing that complex animals existed 580 million years ago would help scientists better understand biodiversity before the Cambrian Period, says Ronald Jenner of the University of Bath in the United Kingdom. If the bilaterians were there, then the “basic branches of the animal kingdom [had] already been established at this point,” Jenner says, as indirect evidence from modern animals’ genes has suggested.

—KATHERINE UNGER