in leaf biomass and might also increase if trees grow taller, which would intensify competition for sunlight. Rapid growth in tree height drives strong responses to nutrient supplementation in young forests recovering from disturbances, but whether this is the case in old-growth tropical forests is not known[8]. This uncertainty is compounded by the fact that long-lived trees that are adapted to infertile soils allocate substantial amounts of carbon and nutrients to storage and defence, and are thus characterized by both slow growth and low mortality rates. Added nutrients might be acquired and stored for many years before a productivity response becomes evident.

Cunha and colleagues report statistically significant responses to nutrient addition that are broadly consistent with these biogeochemical and life-history expectations. There was no significant response to nitrogen addition, whereas phosphorus addition increased the production of leaves and fine roots. The rise in leaf production was associated with increased leaf turnover rates, without an increase in leaf biomass. Thus, extra wood production was not required to support increased leaf biomass, and wood production was similar across all nutrient-addition treatments.

If phosphorus were the only limiting soil resource, fine-root production would be expected to decrease in response to phosphorus addition. The observed increase in fine-root production implicates a second, unidentified limiting soil resource as driving the rise in root production.

Previous work[9] indicates that addition of the three major cations causes notable changes in root-growth dynamics, root characteristics and root colonization by nutrient-supplying fungi called mycorrhizae. This suggests that cation levels also limit plant function and might limit biomass production over a longer timescale. Fundamental questions concerning how tropical forests maintain high levels of biomass productivity despite growing on impoverished soils remain to be explored through this rich ongoing experiment.

Cunha and colleagues' results also have major societal implications. The phosphorus limitation of leaf and fine-root production will limit the acquisition of other resources, thereby lowering forest resilience as local climates change (for example, affecting water uptake during drought). Nonetheless, the phosphorus limitation of the forest carbon sink has not been proved fully. Leaves and fine roots turn over quickly and contribute to long-term carbon sequestration only indirectly, by enabling increases in the biomass of long-lived wood and decomposition-resistant soil organic material that persist on a timescale of decades to centuries[10]. Cunha and co-workers' results are from the first two years of their experiment. Long-lived carbon pools will increase with phosphorus addition as the experiment continues, if decomposition-resistant organic compounds in leaves and roots are added to pools of soil organic matter, and if trees adapted to phosphorus-impoverished soils slowly increase wood production.

A previous compilation of soil phosphorus measurements provides a global perspective on the potential nutrient limitation of the forest carbon sink[11]. Highly weathered, low-phosphorus soils predominate across eastern and central Amazonia, and occur throughout the tropics. However, soil phosphorus concentrations vary 100-fold across humid, lowland tropical forests, and broadly overlap with those in temperate and boreal forests (Fig. 1).

This broad variation in soil phosphorus content, together with previous evidence for both nitrogen and phosphorus limitation in forests at all latitudes[7,8,12,13], suggest that future projections of the forest carbon sink must incorporate nutrient cycles.

Eleven Earth-system models[14], which simulate physical, chemical and biological processes to predict future climates, inform the sixth assessment of the Intergovernmental Panel on Climate Change[15]. Five of these models lack nutrient cycles, and only six include a nitrogen cycle (one of which includes both nitrogen and phosphorus cycles). Of these 11 models, the one with both nitrogen and phosphorus cycles predicts the smallest terrestrial carbon sink[14]. Amazonia comprises approximately 25% of the global forest carbon sink[1], and an understanding of nutrient cycles in characteristic low-phosphorus Amazonian soils will improve predictions of the future global forest carbon sink. Cunha and colleagues' experiment has provided, and should continue to provide, crucial information with which to parameterize and validate Amazonian nitrogen, phosphorus and cation cycles, and thereby improve the accuracy of such projections.

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Particle physics

Evidence at last that the proton has intrinsic charm

Ramona Vogt

An analysis of the distribution of the elementary particles that make up the proton provides evidence that it contains a type of quark known as an intrinsic charm quark — verifying a proposal made four decades ago. **See p.483**

Physics textbooks describe the proton as a subatomic particle that contains three quarks, bound together by elementary particles known as gluons. But quantum theory predicts that the proton can contain other quark–antiquark pairs, including some quarks — known as charm quarks — that are more massive than the proton itself. These charm quarks are thought to be 'intrinsic', meaning that they are part of the proton over long timescales and not produced by interactions with a particle that is external to the proton[1,2]. However, attempts to confirm intrinsic charm in experiments have so far fallen short. On page 483, the NNPDF Collaboration[3] reports an analysis of collision data that acts as evidence — if not the discovery — of intrinsic charm in the proton.

The intrinsic-charm content of the proton is expected to differ from the charm–anticharm quark pairs that are generated by high-energy radiation — for example, when a photon fuses with a gluon in an electron–proton collision.

In such a collision, 'extrinsic' charm quarks appear in the centre of the collision, so the statistical distribution of their position, averaged over many experiments, dies away farther from the centre. Instead, in the intrinsic-charm picture that was first proposed in the early 1980s, the charm quark continues moving along the direction of travel of the parent proton in a collision between, for example, an electron and a proton, or between two protons[1,2]. The intrinsic charm quark is therefore expected to appear in the 'forward region' with respect to the centre of mass of the collision. The distribution of charm quarks in the proton determined by the NNPDF Collaboration's analysis is, in fact, similar to that first predicted four decades ago[1].

At the most basic level, a proton with intrinsic charm would be made up of a charm–anticharm pair, together with two 'up' quarks and one 'down' quark. Theoretically, it should be easy to tell the difference between this intrinsic-charm state and the radiatively generated extrinsic state. In practice, however, charm quarks that are produced in the forward region can be difficult to detect and are thus statistically less likely to be observed than are those that appear in the centre.

In the early 1980s, the European Muon Collaboration measured the charm-quark structure function for the proton, which describes the momentum distribution of charm quarks in the proton[4]. The team found indications that could be attributed to intrinsic charm, but low statistical precision made the result inconclusive. Subsequent experiments in the 1990s extended the range of particle momenta over which the measurements were taken, but it was not possible to fit the whole data set in terms of a single intrinsic-charm scenario, and this was taken as evidence against the idea that the proton contains intrinsic charm[5,6]. However, these measurements were not considered definitive.

The presence of intrinsic charm was also hinted at by other types of data, such as those showing differences in the distributions of particles known as mesons. A proton with intrinsic charm (containing two up quarks, one down quark and a charm–anticharm pair) can make a meson — known as a leading meson — that contains either an up–anticharm pair or a down–anticharm pair. The corresponding antiparticles (antiup–charm or antidown–charm) cannot be made from an intrinsic state and can be generated only through the extrinsic scenario. These mesons are known as non-leading mesons.

Leading mesons are produced in the forward region, and their non-leading counterparts are produced close to the collision centre. Their distributions are therefore different, and this difference is referred to as an asymmetry. Asymmetries that have been detected in the distributions of mesons[7–9] could arise from intrinsic charm in the proton, but there are other models[10] of particle production based on connections between quarks that can explain this discrepancy, so these data cannot be taken as definitive proof of intrinsic charm.

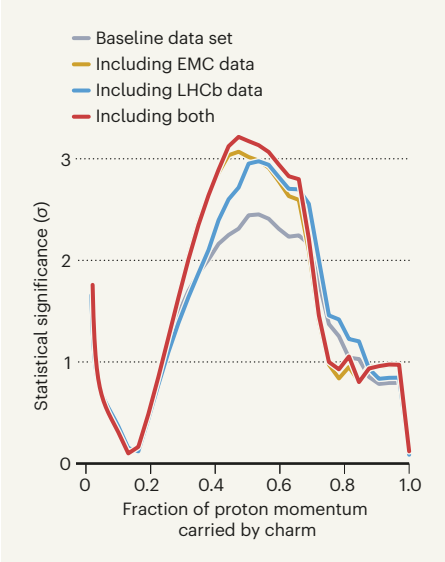

Figure 1 | Evidence of intrinsic charm in the proton. The NNPDF Collaboration[3] used machine-learning techniques to analyse data from experiments designed to determine whether the proton contains an elementary particle, known as a charm quark, that is 'intrinsic' to the proton. The authors found that intrinsic charm is present in the proton with a statistical significance of 2.5 times the standard deviation (σ). The baseline analysis excluded measurements[4] made by the European Muon Collaboration (EMC) in the early 1980s — data that are generally thought to be too imprecise to be conclusive — and those[19] announced in July 2021 by the LHCb collaboration at the Large Hadron Collider at CERN. Including these data in the analysis had the effect of increasing the statistical significance to 3σ, which is considered evidence of an effect in particle physics. (Adapted from Fig. 2 of ref. 3.)

Because no clear evidence had surfaced over several decades, interest in intrinsic charm waxed and waned. However, there has been something of a renaissance in the topic in the past 10 years or so, with several alternative models of intrinsic charm being put forward[11–14]. These various models have been used in analyses[15–18] of data from many different collision experiments. Such analyses have attempted to set limits on the intrinsic-charm content of the proton by assuming that it takes one or all of the forms set out in the models, but, so far, they have returned contradictory or inconclusive results.

The NNPDF Collaboration's study is unique among these analyses in that the authors did not make any assumptions about the way the constituents of the proton are distributed. Instead, they used machine-learning techniques that are agnostic to any particular model. A previous paper[18] by members of the same team found some evidence to support the postulate that intrinsic charm can be detected in the forward region, but this evidence was not definitive. In the present work, the authors distinguished the distributions of momenta that include the charm quark as part of the proton from those that would be expected if charm is generated only radiatively — and found compelling evidence that intrinsic charm is indeed present in the proton.

The authors found that the resulting distribution of charm quarks peaked at a maximum fraction of the proton momentum that is close to that predicted in the 1980s. The effect is small, as expected, because the intrinsic charm carries less than 1% of the total momentum of the proton. However, the result is robust with respect to the mass of the charm quarks, the methodology and the data sets that are included in the analysis. The NNPDF Collaboration concludes that intrinsic charm is present in the proton with a statistical significance of three times the standard deviation (Fig. 1). This is considered evidence of an effect in particle physics — but not a discovery, which is a term reserved for a significance of five times the standard deviation.

In July 2021, data[19] were announced by researchers working at the Large Hadron Collider at CERN, Europe's particle-physics laboratory near Geneva, Switzerland (see go.nature.com/3p2tswa). In these experiments, the particles that were produced in proton–proton collisions seemed to suggest the presence of intrinsic charm in the proton — providing some of the clearest experimental indicators so far for intrinsic charm. Including these data in its analysis helped the NNPDF Collaboration to confirm its results, but its evidence does not rely on these data. Other experiments have been planned at lower energies[20] than those previously used to study intrinsic charm in the proton, and these experiments could provide insight into the conditions under which intrinsic charm is expected to appear.

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Genomics

Genetic variants that edit risk of immune disease

Kaur Alasoo

A process called A-to-I RNA editing helps to prevent unwanted immune responses. Associations between genetic variants and this type of RNA editing now provide mechanistic insights into the genetic basis of autoimmune diseases. **See p.569**

The genetic variants that increase the risk of disease do so by exerting effects on RNA. Variants can alter the amount of RNA produced from a given gene. They can alter the transcribed RNA sequence itself, leading to changes either in the amino-acid sequence of the encoded protein or in how much of the RNA is removed through 'splicing' before it is translated into protein. Finally, variants can influence whether RNA molecules undergo A-to-I RNA editing[1] — a process in which a subunit of RNA is changed from adenosine (A) to inosine (I). On page 569, Li *et al.*[2] present a comprehensive analysis of the effects of genetic variants on A-to-I RNA editing. Their work provides new insights into the mechanisms by which genetic variants mediate the risk of some immune-related diseases.

A-to-I RNA editing disrupts the formation of double-stranded RNA molecules. These molecules occur naturally, either when both a protein-coding gene and its complementary DNA are transcribed and the transcripts form a duplex, or when non-coding, repetitive transcripts form duplexes with themselves. Conversion of A to I by the enzyme ADAR disrupts the RNA duplex. But unedited RNA duplexes are bound by the sensor protein MDA5 which, by mistaking them for viral RNA, triggers an immune reaction against the body's own cells that can lead to autoimmune disease (Fig. 1).

The extent to which variation in RNA editing, mediated by genetic variation, affects the risk of autoimmune diseases has been unclear. Although this question has been touched on before[3], Li and colleagues took a broader approach, studying genomes and RNA sequences from 49 human tissues from 838 people, using data from a project called GTEx (ref. 4). The authors analysed how patterns of RNA editing varied between individuals, to identify *cis* genetic variants (those located close to editing sites) that are associated with RNA editing. These variants are dubbed editing quantitative trait loci (edQTLs).

"The study highlights the importance of considering all possible mechanisms by which a genetic variant might influence disease risk."

The researchers found that the edQTLs are largely different from the variants that regulate gene expression or splicing. They are clustered in specific genomic features, such as regions that encode A-to-I editing sites, and the authors provide evidence that the variants might alter the strength with which ADAR can bind to RNA.

Li *et al.* next examined the overlap between their edQTLs and variants that other studies have associated with autoimmune and immune-mediated diseases. They found that, in general, edQTLs associated with decreased levels of RNA editing confer a higher-than-average risk of autoimmune and immune-mediated diseases. This directionality contrasts with the behaviour of risk variants for these diseases that alter gene expression and splicing. Such variants can either increase or decrease protein function, depending on the part that the protein plays in the disease.

Finally, the authors zoomed in to identify specific edQTLs that bring about disease-associated changes in editing, and found that 33% of these genomic regions encode proteins. It is intriguing to think that, at these loci, it might not be changes in the function of the protein that contributes to disease risk (as is often assumed to be the case), but rather the formation of double-stranded RNA that leads to activation of the innate immune system. The function of the encoded protein might even be completely irrelevant for the disease.

However, individual genetic variants often have effects on multiple genes, as well as on different layers of regulation (that is, on expression, splicing and editing). Although the authors used careful analysis to try to reduce such confounding factors, they cannot be completely avoided. For example, around 20% of the genetic variants associated with changes in RNA editing were also associated with altered gene-expression levels or splicing. This means that, for any given locus, it is difficult

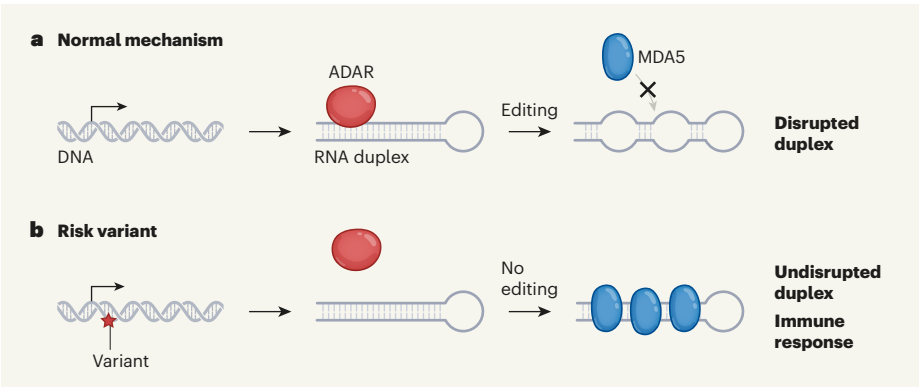

Figure 1 | Varying risk for autoimmune disease. a, Some regions of the genome give rise to RNA that forms a double-stranded duplex. This RNA can be edited by the enzyme ADAR, which converts a subunit of the RNA called adenosine into inosine (not shown), disrupting the duplex and preventing the viral-sensor protein MDA5 from binding. **b**, Li *et al.*[2] provide evidence that some genetic variants are associated with lower levels of this type of RNA editing, perhaps because ADAR does not bind the RNA so strongly. Unedited RNA duplexes are bound by MDA5, which wrongly recognizes the RNA as 'non-self' and triggers immune responses. Thus, these genetic variants are also associated with a higher risk of developing some immune-related diseases.