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Decoding abiotic stress resilience in *Solanum*: Transcriptional and epigenetic landscapes

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The environment proposes, the organism disposes

(Georges Canguilhem)

INDEX

ABSTRACT

Plant resilience to abiotic stress represents a critical challenge for modern agriculture, as conditions such as drought and salinity severely compromise crop productivity in an increasingly unstable climate. Potato (*Solanum tuberosum* L.), one of the world's most important food crops, provides a valuable model for investigating the biological mechanisms underlying stress adaptation. Its wild relative, *Solanum commersonii*, however, exhibits remarkable natural tolerance and offers a valuable system for understanding the genetic and regulatory factors that sustain resilience. In plant stress responses, an important role is played both by secondary metabolites, including anthocyanins involved in cellular protection, and by epigenetic mechanisms that modulate gene expression and contribute to adaptive plasticity. Integrating these levels is therefore central to deciphering plant resilience. Following an introductory chapter, the Chapter II examines the response of cultivated potato (*S. tuberosum*) to drought stress through a comparison of four genotypes with differing pigmentation (two anthocyanin-rich and two non-pigmented). The plants were subjected to six days of water stress, followed by a rewatering phase on the seventh day. The results indicate that resilience is closely linked to the constitutive (preventive) accumulation of anthocyanins rather than their stress-induced synthesis. Molecular analysis reveals that a pre-existing pool of these pigments allows for a more flexible transcriptional response, mediated by the coordinated regulation of key genes such as *DFR*, *AN1*, and *AN2*, thereby optimizing oxidative protection. The Chapter III addresses an interspecific comparison between *S. tuberosum* (cv. Désirée) and the wild species *S. commersonii* (clone 1T) under prolonged salt stress (21 days), involving three increasing levels of salinity (0, 80, and 150 mM NaCl). The data highlight that the superior tolerance of *S. commersonii* appears to derive from greater regulatory plasticity sustained by targeted chromatin remodeling (gain of H3K4me3 and loss of H3K27me3) at key genes for antioxidant defense and phenylpropanoid metabolism, supported by ChIP-qPCR and complemented by qualitative locus-level ChIP-seq profiles (highest-quality replicate retained due to mapping constraints). Furthermore, the identification of the specific microRNA miRNA19 suggests a resource reallocation strategy toward defense, mediated by the repression of cell growth programs. The Chapter IV addresses heat stress and extends the investigation to heat stress memory (priming) in tomato (*S. lycopersicum*). The study demonstrates that priming establishes a transcriptional memory characterized by the anticipatory activation of thermotolerance-related pathways, including those mediated by heat shock proteins (HSPs) and chloroplast

function. The results suggest that stress memory represents an additional layer of adaptation that can be exploited in combination with the use of wild relatives to develop potato cultivars with improved resistance to recurrent environmental stresses. Overall, this work demonstrates that enhanced stress tolerance emerges from the interplay between efficient metabolic pathways, dynamic transcriptional regulation, and advanced epigenetic control, providing new perspectives for breeding strategies aimed at improving crop resilience.

CHAPTER I

General introduction

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Introduction

1.1 The potato (*Solanum tuberosum*) and its wild relatives

The Solanaceae family represents one of the largest and most diverse groups of angiosperms, comprising about 3,000 species distributed across approximately 100 genera (Olmstead et al., 2008). The highest concentration of diversity within this family is found in Central and South America, which represent the center of origin for many of its species. This family includes several crops of major agricultural and nutritional importance, such as tomato (*Solanum lycopersicum*), potato (*S. tuberosum*), and eggplant (*S. melongena*), as well as peppers (*Capsicum spp*.), ground cherries (*Physalis spp*.), and tobacco (*Nicotiana spp*.) (Motti, 2021). Widely cultivated ornamental species such as Petunia, Datura, and Atropa also belong to this family. In addition, several Solanaceae species, including tomato, tobacco, and petunia, serve as key biological models for studying plant development, stress responses, and hormonal regulation (Gebhardt, 2016). Thanks to their remarkable diversity, Solanaceae species provide an ideal model for comparative genomics, allowing the exploration of a wide range of genetic traits and evolutionary adaptations.

Within this framework, the potato (*S. tuberosum*) is the most important Solanaceae worldwide and it is recognized as the first non-cereal crop in the world (it ranks fourth among staple foods after rice, wheat, and maize, FAO, 2021) in terms of production value, caloric contribution, and global food security. Its domestication began around 8000–5000 BC in the Andes region of South America, particularly around Lake Titicaca, which lies at an elevation of 3,800 meters (12,500 feet) and forms the border between Bolivia and Peru. It is believed that humans began consuming wild potatoes from the humid coastal plains of South America about 13,000 years ago. Today, potato is a cornerstone of global agriculture. According to FAO data, global potato production reached approximately 383 million tonnes in 2023, cultivated over an area of around 16.8 million hectares (FAOSTAT, 2024) (Figure 1B). This high output, combined with the crop's short growing cycle and adaptability to diverse environments, contributes to its strategic role in global food security. The potato is not only consumed fresh but is also extensively used in processed food production. Moreover, it serves as an industrial source of starch and alcoholic beverages (Reddy et al., 2018). Potato tubers represent an important source of complex carbohydrates, fibers, vitamins, and bioactive compounds, including secondary metabolites of nutraceutical interest such as anthocyanins and other flavonoids

(Barbas et al., 2023). Its extensive cultivation across both developing and industrialized regions makes it particularly valuable for addressing the food needs of growing populations and supporting economic development (Varga & Đurović, 2023).

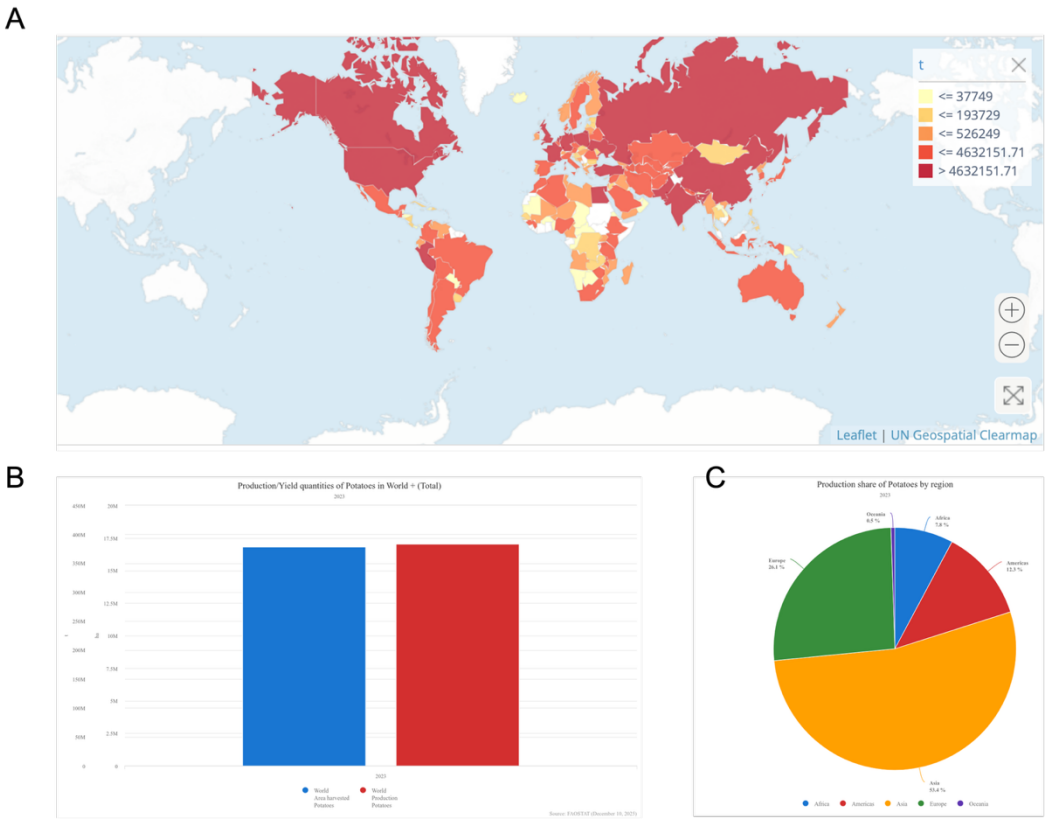

Figure 1. Global distribution, productivity, and regional contribution of potato production in 2023. The upper panel shows a world map of potato production based on FAOSTAT 2023 data, highlighting major producing areas. The lower-left panel compares global harvested area and total production, illustrating overall productivity. The lower-right panel presents the regional share of global potato output, with Asia accounting for the largest proportion (FAOSTAT 2024).

Alongside the cultivated species, wild potato relatives within the genus *Solanum* represent a valuable genetic reservoir. They possess adaptive traits that have evolved in extreme or marginal environments, making them a key resource for breeding programs (Hawkes, 1990; Jansky, 2010). Many wild species display remarkable tolerance to abiotic stresses such as drought, salinity, and high temperatures, as well as resistance to various biotic stresses. Their characterization and utilization help broaden the genetic base of cultivated potato, which is often limited by high genetic uniformity and reduced adaptability (Bradshaw & Bonierbale, 2010; Spooner et al., 2014). For example, *S. chacoense* and *S. berthaultii* exhibit high heat tolerance, making them valuable genetic resources for breeding thermotolerant cultivars (Smillie et al., 1983). Additional evidence

for drought tolerance in wild *Solanum* comes from recent work showing that the wild species *S. sisymbriifolium* exhibits significantly better growth, biomass accumulation, and stomatal regulation under water stress compared to cultivated species (Kégbé et al., 2024). Among the wild relatives of cultivated potato, *S. commersonii* (Figure 2) is one of the most extensively studied and well-characterized species.) It is a diploid (2n = 2x = 24), tuber-bearing wild potato species belonging to the tertiary gene pool (1EBN), which is sexually isolated from cultivated potato but can be hybridized through unreduced gametes and bridge crosses (Jansky, 2006; Carputo et al., 1997). It is a wild, tuber-producing potato species native to Central and South America. More specifically, *S. commersonii* is endemic to the coastal regions of Argentina, Uruguay, and southern Brazil, where it grows across a wide range of environments, including marshy areas, riverbanks, grasslands, and sandy soils (Hawkes, 1990; Aversano et al., 2021). It exhibits several resistance traits absent in the cultivated potato, including resistance to root-knot nematodes, soft rot, blackleg, bacterial and Verticillium wilt, Potato virus X, tobacco etch virus, common scab, and late blight. Particularly noteworthy is its tolerance to frost and its ability to cold-acclimate (i.e., to increase its freezing tolerance after exposure to non-freezing low temperatures). In contrast, the cultivated potato is classified as frost-sensitive and unable to cold-acclimate (Palta and Simon, 1993). Because of this broad resistance spectrum, *S. commersonii* has long been recognized as an important donor of agronomically valuable traits for potato improvement programs (Micheletto et al., 2000; Gonzalez et al., 2013). The importance of *S. commersonii* has been further reinforced by the fact that it was the first wild potato species whose genome was sequenced, providing fundamental insights into genome evolution, stress adaptation, and the genetic basis of biotic and abiotic stress tolerance in wild potato relatives (Aversano et al., 2015).

Figure 2. Morphological features of *Solanum commersonii*. Images showing the flower morphology with its characteristic purple corolla and yellow anthers, the root system and overall plant and the appearance of plants exposed to 150 mM NaCl salt stress.

Today's agronomic and environmental challenges make the study and use of these genetic resources increasingly urgent, particularly in the context of climate change, which is expected to intensify drought, heat and salinity in many potato-growing regions. In this context, the cultivated potato is particularly vulnerable, showing high sensitivity to these stress factors (Levy & Veilleux, 2007). As a "cool-climate" crop with relatively shallow roots and high-water demand, it is particularly vulnerable to fluctuations in water availability and temperature. Understanding how plants in general perceive and respond to these abiotic stresses at both the physiological and molecular level, including the protective role of secondary metabolites and the contribution of epigenetic regulation, is therefore essential for interpreting stress responses in potato and for exploiting the adaptive traits present in wild potato relatives. In this framework, the following sections outline the core mechanisms underlying plant responses to drought, salinity and heat,

providing the conceptual background for the stress-specific experimental chapters of this thesis.

1.2 Abiotic stresses: physiological responses and core molecular mechanisms

Abiotic stresses are among the main limiting factors for crop growth and productivity. Among these, salinity, drought, and high temperatures are particularly critical, leading to significant yield losses and threatening global food security (Boyer, 1982; Lesk et al., 2016). In the context of climate change and rapid human population growth, the impact of these stresses is expected to become even more severe. The world's population currently stands at 8 billion. According to the latest United Nations projections, this number could increase to about 8.5 billion by 2030 and 9.7 billion by 2050 (United Nations, 2023). Thus, meeting the rapidly growing global food demand represents a major challenge for agriculture under increasingly variable environmental conditions. Climate change and unsustainable agricultural practices are also intensifying key abiotic constraints: soil salinization (driven by factors such as intensive irrigation and seawater intrusion) reduces crop productivity, prolonged drought events limit water availability for agriculture, and more frequent heatwaves strongly affect plant physiological processes (Munns & Tester, 2008; Lesk et al., 2016; Hasanuzzaman et al., 2013). Many aspects of plant physiology are affected by abiotic stress conditions, which cause profound alterations in cellular processes. Some of these changes are non-adaptive and merely reflect stress-induced damage, such as alterations in membrane fluidity or protein structure caused by heat or cold, or disruptions in enzyme kinetics and molecular interactions due to toxic ions. Conversely, other changes are adaptive and enhance plant tolerance, including the repair of stress-induced damage, restoration of cellular homeostasis, and regulation of growth to adjust to new environmental conditions. Consequently, these mechanisms and the molecular networks that control them represent potential targets for genetic improvement of crop species (Zhang, 2020).

In the following subsections, salinity, drought and heat stress are examined in more detail, with a focus on their general physiological effects and on the main molecular signaling and regulatory pathways involved in plant adaptation.

1.2.1 Salt stress

Salinity represents a growing threat, particularly in arid and semi-arid regions. Soil salinity can be classified into two main categories: primary and secondary. Primary salinity develops naturally over long periods as a result of geological and atmospheric processes. Weathering of parent materials containing soluble salts releases sodium, calcium, and magnesium chlorides, together with sulfates and carbonates, with sodium chloride being the most soluble compound. In addition, so-called "cyclic salts," consisting mainly of ocean-derived sodium chloride, are carried inland by wind and deposited on the soil through precipitation (Zhu, 2001). In contrast, secondary salinity is induced by human activities and today represents the most widespread form of agricultural salinization. It results from practices that disrupt the soil's hydrological balance between the water supplied (by rainfall or irrigation) and that actually used by crops through transpiration. The main contributing factors include deforestation and the replacement of perennial vegetation with annual crops, which reduce the soil's ability to regulate the water cycle, as well as irrigation systems that use saline water or have poor drainage, leading to salt accumulation in the rhizosphere (Szabolcs, 1974; Garg and Manchanda, 2008; Shanker and Venkateswarlu, 2011). Excess salts cause osmotic imbalances that reduce water uptake and lead to toxic accumulation of ions, mainly sodium and chloride, thereby impairing enzyme activity and membrane stability. Moreover, salinity stress induces an increase in reactive oxygen species (ROS) production, causing oxidative damage to lipids, proteins, and DNA (Zhu, 2001). High concentrations of Na^+ and Cl^- in the soil thus cause both osmotic and ionic stress, reducing the plant's ability to absorb water and nutrients (Ismail et al., 2020; Ismail, 2014), and salt responses typically show an early osmotic phase followed by a slower ionic phase (Munns and Tester, 2008). The root is the first organ to come into direct contact with salt. Salinity stress significantly reduces root biomass and alters the distribution of components within the root system architecture, affecting the growth rate of both primary and lateral roots and inhibiting their formation. To cope with these adverse conditions, plants activate a variety of physiological and biochemical responses aimed at maintaining ionic homeostasis, proper osmotic potential, and the balance of ROS (Yang et al., 2018).

At the molecular level, salinity is perceived by plants as a combination of osmotic and ionic stress, leading to rapid increases in cytosolic calcium (Ca^{2+}) and ROS, as well as the activation of hormone signaling pathways, particularly abscisic acid (ABA) (Zhu, 2001; Deinlein et al., 2014). These early signaling events activate conserved ion regulatory

mechanisms, most notably the Salt Overly Sensitive (SOS) pathway, which plays a central role in Na^+ extrusion and ionic homeostasis. In this pathway, the Ca^{2+} sensor Salt Overly Sensitive 3 (*SOS3*), also known as calcineurin B-like protein 4 (*CBL4*), and the protein kinase SOS2 (Salt Overly Sensitive 2, also known as *CIPK24*) interact to activate the plasma membrane Na^+/H^+ antiporter *SOS1*, reducing cytosolic Na^+ levels and contributing to K^+/Na^+ homeostasis (Zhu, 2002; Deinlein et al., 2014).

In addition to extrusion mechanisms, plants use vacuolar NHX-type Na^+/H^+ antiporters to sequester Na^+ into vacuoles, reducing cytotoxic effects (Blumwald, 2000). The HKT (high-affinity K^+ transporter) family contributes to salt tolerance by retrieving Na^+ from the xylem stream and limiting its accumulation in the shoot, which is essential for protecting photosynthetically active tissues (Hauser & Horie, 2010; Almeida et al., 2013). These transport processes are tightly regulated by Ca^{2+}-dependent protein kinases and CBL–CIPK signaling complexes, which integrate ionic and osmotic stress signals (Deinlein et al., 2014).

Salinity also triggers transcriptional reprogramming via stress-responsive transcription factors such as NAC (NAM, ATAF1/2 and CUC2), MYB (myeloblastosis), WRKY (WRKY-type zinc-finger proteins) and AP2/ERF (APETALA2/ethylene-responsive factor) and DREB (dehydration-responsive element-binding) proteins, which regulate genes involved in osmoprotectant biosynthesis, ROS detoxification, membrane stabilization, and stress-protective proteins (Golldack et al., 2014; Ismail & Horie, 2017). These changes help maintain osmotic balance and minimize oxidative damage under saline conditions. Importantly, epigenetic mechanisms have emerged as key modulators of salt stress responses, involving DNA methylation, histone modifications, chromatin remodeling, and non-coding RNAs, which together influence gene expression, stress memory, and transgenerational tolerance (Singroha et al., 2022; Zhang et al., 2024; Arıkan et al., 2025). This regulatory landscape will be discussed in more detail in paragraph 1.5.

1.2.2 Drought stress

Drought is one of the most impactful stresses for potatoes, a crop with a relatively high-water requirement. It causes loss of cellular turgor, stomatal closure, and reduced photosynthesis, leading to direct effects on growth and biomass accumulation. At the metabolic level, drought induces the accumulation of osmoprotectants such as proline and soluble sugars and activates antioxidant mechanisms to counteract excessive ROS

production (Chaves et al., 2003; Verslues et al., 2006). Water stress occurs when water loss from the plant system exceeds water uptake, resulting in reduced tissue water content, loss of turgor, decreased cell expansion, and disruption of various physiological processes. The first plant response to drought is a reduction in growth. Lower growth and productivity under drought conditions are mainly caused by altered plant water relations, decreased photosynthetic activity, oxidative stress, membrane damage, and, in some cases, inhibition of enzymatic activity (Batool et al., 2020). The degree of drought tolerance varies among plant species and cultivars, depending on their phenological stage and the duration of stress exposure (Ozturk et al., 2021). Initially, plants respond through morphological adjustments involving stomatal regulation, reducing both stomatal opening duration and aperture size to limit water loss. As water stress persists, photosynthetic damage occurs, which can become lethal under severe conditions. Under these damaging conditions, plants focus on maintaining osmotic adjustment, which is achieved through the biosynthesis and accumulation of osmoprotectants or osmolytes such as soluble proteins, sugars, and alcohols. It is important to note that the synthesis and accumulation of osmolytes are essential for maintaining lower water potentials and minimizing damage caused by water deficit in plants (Ozturk et al., 2021). Drought stress induces extensive transcriptional reprogramming involving multiple gene networks. Microarray and transcriptome analyses show that drought-tolerant genotypes upregulate mitochondrial metabolism while repressing photosynthesis, alongside broad activation of transcription factors, kinases, redox genes, and osmotic adjustment pathways (Evers et al., 2010; Zhang et al., 2014; Yang et al., 2019). Among the regulatory mechanisms, abscisic acid (ABA) plays a pivotal role in coordinating drought-responsive gene expression: activation of the PYR/PYL/RCAR–SnRK2 module induces *ABF*, *ABI5*, WRKY, and HSP transcriptional programs, while ABA-dependent induction of *DREB1A* further enhances tolerance (Bhargava & Sawant, 2013; Chen et al., 2021; Muñiz García et al., 2012). ABA-independent pathways also contribute, with MYB/MYC and WRKY regulators controlling drought-inducible genes independently of ABA (Abe et al., 1997) and NAC transcription factors integrating signals across both pathways (Hu et al., 2006).

1.2.3 Heat stress

Global warming has severely affected crop growth and yield, posing a serious threat to agricultural production and food security. By the middle of the 21st century, the annual maximum daily extreme temperature is expected to rise by approximately 1–3 °C (Field et al., 2012). High temperatures compromise membrane stability, protein functionality, and enzymatic activity. Protein denaturation reduces the efficiency of biochemical processes, while membrane instability increases electrolyte leakage and tissue dehydration. Plants respond by producing heat shock proteins (HSPs) and activating antioxidant systems to mitigate oxidative stress (Kotak et al., 2007; Mittler et al., 2012). Plants growing at temperatures above their optimal range for extended periods experience heat stress (Wahid et al., 2007). This stress can cause irreversible damage to plant growth, significantly reduce yield and quality in major food crops, and even lead to plant death (Fahad et al., 2017; Ni et al., 2018; Zafar et al., 2018). At the molecular level, photosynthetic efficiency is easily impaired by heat stress, leading to a shortened life cycle and reduced yield. Respiratory enzyme activities decline, resulting in energy shortages, while heat-induced movement of biomolecules disrupts plasma membrane homeostasis, severely altering its permeability and fluidity and causing ion and amino acid leakage. In general, when the optimal growth temperature of a plant is exceeded by 5–10 °C, a burst of reactive oxygen species (ROS) occurs, leading to irreversible oxidative damage at the cellular level, particularly in photosystems I and II (Pucciariello et al., 2012; Zhao et al., 2020). Several studies in *Arabidopsis thaliana* have revealed a highly structured transcriptional network underlying the heat stress response (HSR). At the top of this hierarchy are the HsfA1 transcription factors, which function as master regulators indispensable for activating heat-responsive gene expression. Functional studies in tomato and *Arabidopsis*have shown that knockdown or knockout of *HsfA1* genes severely compromises the induction of heat-responsive genes and leads to strong heat-sensitive phenotypes (Mishra et al., 2002; Yoshida et al., 2011; Liu et al., 2011). HsfA1 factors directly regulate several core heat-responsive transcription factors, including *DREB2A*, *HsfA2*, *HsfA7a*, *HsfBs*, and *MBF1C*, thereby orchestrating the early phases of the HSR (Yoshida et al., 2011). Among these, *HsfA2* acts as a major amplifier of the heat stress response. Plants lacking *HsfA2* exhibit pronounced heat sensitivity and reduced expression of numerous HS-inducible genes, demonstrating its essential role in sustaining thermotolerance (Charng et al., 2007). Heat stress also generates a splicing variant of *HsfA2* (*S-HsfA2*) that binds to and activates its own promoter,

reinforcing *HsfA2* induction through a positive autoregulatory loop (Liu et al., 2013). Together, these regulatory interactions define a hierarchical and dynamic transcriptional architecture that ensures precise modulation of plant heat stress responses.

1.3 *Solanum tuberosum* and abiotic stress

The cultivated potato is notoriously sensitive to salinity, drought, and heat, which greatly reduce both yield and tuber quality. Heat stress is considered the main environmental factor affecting potato growth, tuber yield, and quality (Levy et al., 2007). High temperatures lead to reduced photosynthetic capacity and starch accumulation as a result of inhibited carbon transport, CO_2 fixation, and chlorophyll degradation (Hancock et al., 2014). In addition to limiting the plant's metabolic efficiency, heat stress also negatively affects tuber quality traits, such as skin appearance (Molteberg et al., 2017). In contrast, several wild Solanum species exhibit greater adaptive capacity, reflecting evolutionary strategies developed in harsh environments (Carputo et al., 2003; Obidiegwu et al., 2015). Recent transcriptomic studies have begun to dissect the molecular basis of these responses in potato, revealing regulatory networks of ERF and deubiquitinase (DUB) genes involved in integrated responses to drought and heat (Li et al., 2024).

Among the various abiotic stresses that compromise potato growth and productivity, salinity stress is one of the most critical and damaging. In potato, high salt concentrations in the soil cause a marked reduction in vegetative growth and yield. It has been reported that under treatment with 50 mM NaCl, plant growth decreases and tuber production is reduced by about 50%, while at 150 mM NaCl, growth is completely inhibited (Hmida-Sayari et al., 2005). As discussed for plants in general, salinity in potato causes osmotic and ionic stress, leading to nutrient imbalances and membrane damage, with a consequent reduction in photosynthetic activity and efficiency of detoxification mechanisms against ROS. During the early stages of salt exposure, the physiological response involves stomatal closure and reduced leaf expansion; subsequently, the accumulation of toxic ions in older leaves induces premature senescence, reducing photosynthetic capacity (the "source") and ultimately leading to leaf tissue death (Gholizadeh, 2017). In line with these observations, Efimova et al. (2018) showed that, under salinity conditions, plants of the cultivar Lugovskoi exhibit strong inhibition of growth processes, reduced chlorophyll a content, and suppression of stolon formation, indicating a low level of salt tolerance. Conversely, wild potato species generally display greater tolerance to abiotic stresses. However, intensive selection for high yield, tuber storability, pathogen resistance, and

optimal maturation time has progressively reduced salt tolerance in modern cultivars compared to their wild relatives. Another major environmental factor affecting potato productivity is drought, to which this species is highly sensitive, mainly due to its shallow root system, which limits access to deeper water reserves (Obidiegwu et al., 2015). The effects of drought on potato growth and development vary greatly depending on the specific canopy and root characteristics of each cultivar, as well as on abiotic factors such as stress duration, onset time (Jefferies and MacKerron, 1993), and severity (Stark et al., 2013). Other elements, including soil moisture, nutrient availability (Saravia et al., 2016), and atmospheric evaporative demand (Jefferies and MacKerron, 1993), further influence plant responses. The potato's developmental cycle comprises five main phenological stages: plant establishment, stolon initiation, tuber initiation, tuber bulking, and maturation (Obidiegwu et al., 2015). Water stress can reduce yield during several of these stages by affecting vegetative growth through reduced plant height, leaf number, and leaf size (Deblonde et al., 2001) and by decreasing photosynthetic capacity through reduction in chlorophyll content, leaf area index, and photosynthetic activity duration. Recent observations have also highlighted that deeper or denser root systems within moist soil layers improve water-use efficiency, suggesting morpho-functional traits that could be targeted in potato breeding (Chen et al., 2022). Beyond compromising vegetative growth, drought can also negatively affect the reproductive phase, shortening the growth cycle (Kumar et al., 2007) or reducing both the size (Schafleitner et al., 2009) and number of tubers produced (Eiasu et al., 2007). Moreover, water stress can worsen tuber quality by altering starch content and other compositional traits (Ekanayake et al., 1989). Recent studies have also shown that the combination of multiple stresses, such as drought and saline-alkaline conditions, can have a synergistically detrimental effect, leading to more than a 50% reduction in tuber yield over two consecutive growing seasons (Zhang et al., 2022).

1.4 Shared adaptive responses to abiotic stress

Despite the differences among the three types of stress, some adaptive responses are shared: the synthesis of osmoprotectants, the activation of antioxidant enzymes and metabolites, including anthocyanins, and the regulation of hormones such as abscisic acid (ABA) and jasmonates represent key mechanisms for maintaining cellular homeostasis (Cutler et al., 2010). In this context, pathways of secondary metabolism, particularly the flavonoid and anthocyanin branches, play a central role in stress tolerance. In the

following section, particular attention is given to the role of secondary metabolites, especially anthocyanins, as key components linking environmental cues to plant stress responses.

1.4.1 Anthocyanins and secondary metabolism in plant stress responses

Anthocyanins are water-soluble pigments belonging to the flavonoid class, responsible for the red, purple, and blue hues of many plant organs. Beyond their aesthetic and ecological roles, related to pollinator attraction and seed dispersal, these molecules play essential protective functions in plants (Gould, 2004). They act as powerful antioxidants, contributing to the detoxification of reactive oxygen species (ROS), protecting tissues from ultraviolet-induced damage, and participating in the mechanisms of adaptation to environmental stresses (Chalker-Scott, 1999; Winkel-Shirley, 2002). At the same time, the growing interest in anthocyanins also stems from their nutraceutical properties, which make them compounds of great relevance for human health and industrial applications (He & Giusti, 2010).

Anthocyanin biosynthesis is part of the broader phenylpropanoid pathway. Within the flavonoid biosynthetic pathway, related metabolites such as proanthocyanidins are also synthesized from epicatechin via anthocyanidin reductase (*ANR*), further expanding the range of phenolic compounds involved in plant defence (Villegas et al., 2016). This pathway involves numerous genes, which can be schematically divided into structural, modifying, and regulatory genes. The structural genes located upstream in the pathway, such as phenylalanine ammonia-lyase (*PAL*) and 4-coumarate-CoA ligase (*4CL*), are later classified as early biosynthetic genes (EBGs) and late biosynthetic genes (LBGs). The early biosynthetic genes include chalcone synthase (*CHS*), a key enzyme in the pathway, chalcone isomerase (*CHI*), flavanone 3 hydroxylase (*F3H*), and flavonoid 3',5' hydroxylase (F3'5'H). The LBGs are further divided into biosynthetic and modifying genes. The biosynthetic LBGs comprise dihydroflavonol 4 reductase (*DFR*), anthocyanidin synthase/leucoanthocyanidin dioxygenase (ANS/LDOX), and UDP glucose flavonoid glucosyltransferase (*UFGT*). The modifying genes include methyltransferases (*MT*), O-methyltransferases (*OMT*), and anthocyanin acyltransferases (*AT*) (Figure 3).

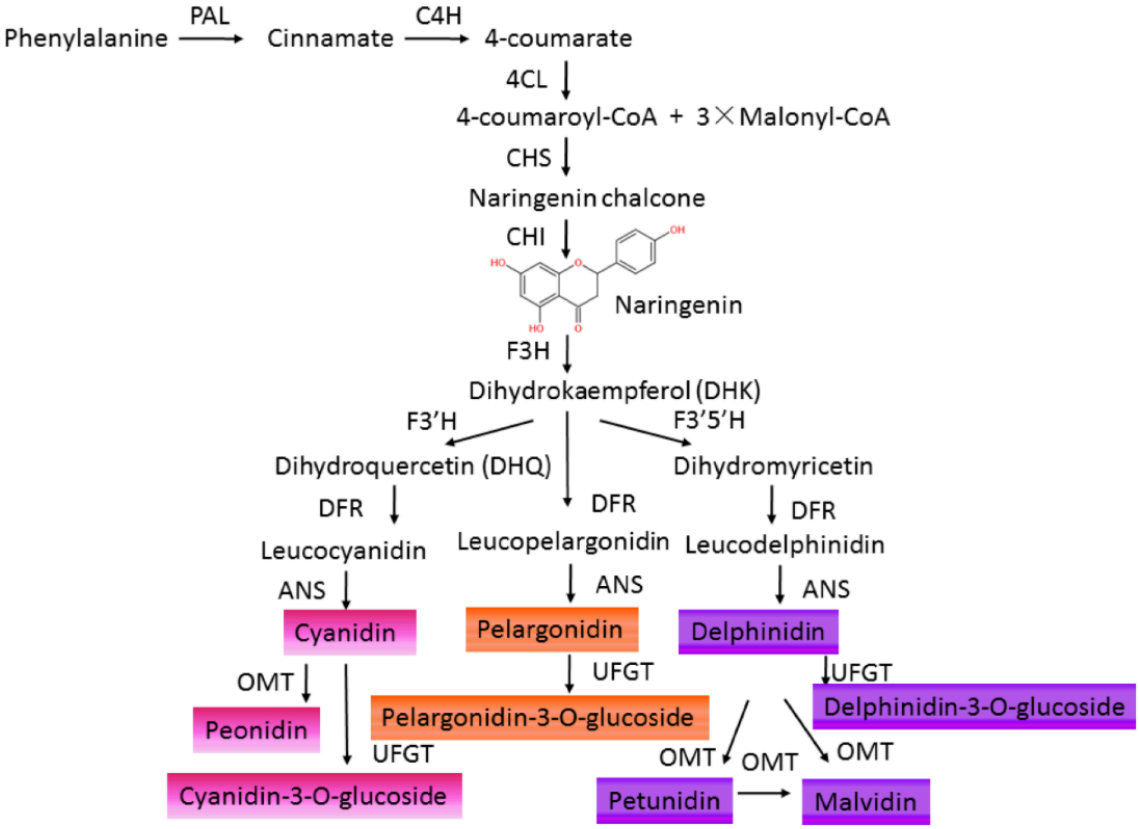

Figure 3. The pathway of anthocyanin biosynthesis. PAL, phenylalanine ammonia lyase; C4H, cinnamate 4-hydroxylase; 4CL, 4-coumarate CoA ligase; CHS, chalcone synthase; CHI, chalcone isomerase; F3H, flavanone 3-hydroxylase; F3′H, flavonoid 30 hydroxylase; F3′5′H, flavonoid 3050hydroxylase; FLS, flavonol synthase; DFR, dihydroflavonol 4-reductase; ANS, anthocyanidin synthase; UFGT, UDP-galactose flavonoid 3-O-galactosyltransferase; OMT, O-methyl transferase. (Yanyun Ma et al., 2021).

The entire biosynthetic pathway is regulated by the transcription factor genes MYB, bHLH, and WD40, which modulate the expression of structural genes. These transcription factors assemble into a single regulatory complex known as MYB–bHLH–WD40 (MBW) complex, enhancing their combined effect (Goswami et al., 2018). The expression of MYB genes, mainly belonging to the R2R3 MYB family, and bHLH genes is largely specific to pigmented tissues. In contrast, the expression of WD40 genes, which are involved in stabilizing the MBW complex, is similar in both pigmented and non-pigmented tissues (Koes et al., 2005). The MYB, bHLH, and WD40 transcription factors regulate the expression of late structural genes by binding to specific cis-acting elements in their promoter regions. The MBW complex is formed through the interaction of MYB, which is influenced by both evolutionary and environmental factors, with the N-terminal 200 amino acids of bHLH, which in turn interacts with the subsequent 200 amino acids of WD40. The MBW complex is further divided into MREs (MYB Recognition

Elements) and BREs (bHLH Recognition Elements), which bind to the promoter region of the ta

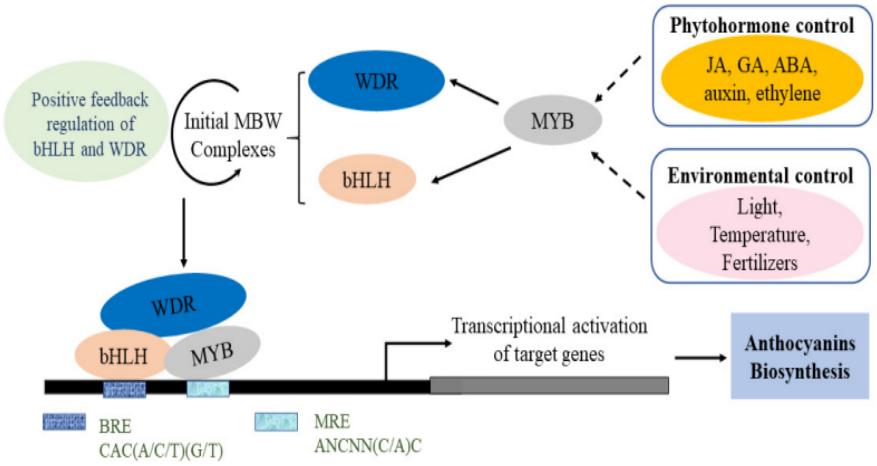

Figure 4. Simplified model of the action of the MBW complex in anthocyanin biosynthesis (Huimin Liu et al., 2021).

Beyond their developmental and ecological roles, anthocyanins and other flavonoids are strongly inducible under abiotic stress. Several studies have shown that anthocyanin biosynthesis is strongly influenced by environmental conditions. Abiotic stresses such as salinity, drought, and heat can induce an increase in the accumulation of these pigments, which act as integral components of the plant's defense strategies (Nakabayashi et al., 2014). Although anthocyanins are secondary metabolites, they are classified as specialized plant metabolites (SPMs) because of their central protective role in stress adaptation. In response to abiotic stress, the content of polyphenols and flavonoids increases, promoting plant adaptation to unfavorable environments (Sharma et al., 2019). It has been observed that the flavonoid content increases under drought stress in wheat cultivars (Ma et al., 2014). In *Arabidopsis thaliana*, Serrano et al. (2019) reported a strong increase in the flavonoid naringenin. Anthocyanins are often overexpressed upon exposure to cold, compensating for the reduced protective effect of ascorbic acid by performing an antioxidant function (Xie et al., 2012). In apple, for example, cold stress enhances the expression of the *DFR* and *UFGT* genes in the flavonoid biosynthetic pathway (Zhou et al., 2020). Prolonged exposure to light has also been reported to affect anthocyanin levels (Ban et al., 2007), and these pigments also serve as a defense mechanism against UV-B radiation (Isah and Yadav, 2019). A recent review (Sun et al., 2023) has highlighted that anthocyanin accumulation represents a crucial adaptive trait under drought and salinity, contributing to ROS scavenging and membrane stabilization.

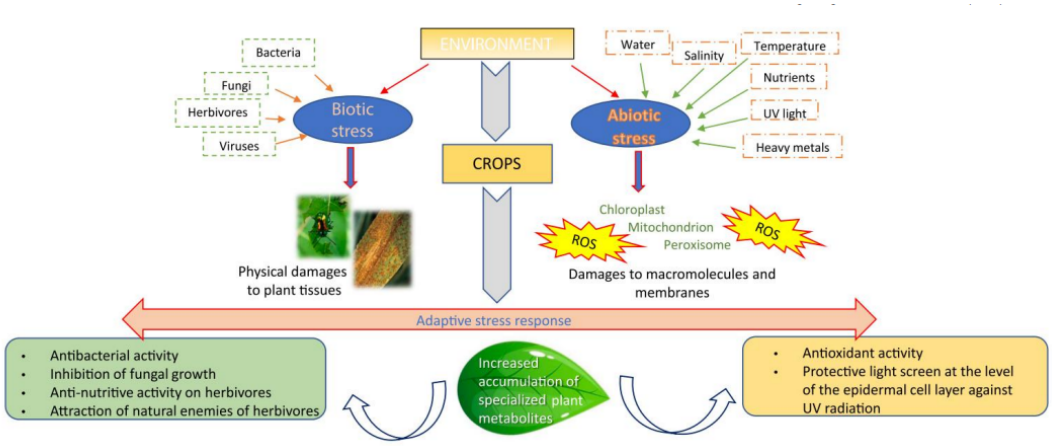

Figure 5. Schematic representation of plant responses to abiotic and biotic stresses.

Taken together, these observations support a model in which the phenylpropanoid and flavonoid pathways, regulated by the MBW complex and other transcriptional networks, integrate environmental cues and contribute to antioxidant defense, membrane protection and overall stress tolerance. This general framework provides the basis for understanding species-specific roles of anthocyanins, including those in potato.

1.4.2 Anthocyanins in potato: composition and regulation

Potatoes are an important source of dietary energy, containing not only primary metabolites (carbohydrates) but also a wide variety of secondary metabolites, among which phenolic compounds play a significant role. As a staple food for the global population and a primary dietary component for certain ethnic groups, the potato has increasingly become the focus of research over recent decades, with several agronomic and genetic improvement strategies explored for its biofortification (De Lapeleire et al., 2018). Unlike other Solanaceae species, such as eggplant (*S. melongena*) and tomato (*S. lycopersicum*), the potato contains all six major anthocyanins (Pg, Cy, Dp, Pn, Pt, and Mv), which contribute to its wide phenotypic diversity. In red-fleshed tubers, pelargonidin-3-(p-coumaroylrutinoside)-5-glucoside is the predominant anthocyanin, while lower levels of peonidin-3-(p-coumaroylrutinoside)-5-glucoside and pelargonidin-3-(trans-feruloyl-rutinoside)-5-glucoside are present. In purple-fleshed tubers, petunidin-3-(p-coumaroyl-rutinoside)-5-glucoside is the main anthocyanin. Additionally, malvidin, peonidin, and delphinidin 3-(p-coumaroylrutinoside)-5-glucosides have been identified in different purple-fleshed cultivars (Lachman et al., 2009). The main anthocyanins in colored-flesh potatoes are derived from the 3,5-diglucosides of cyanidin and, to a lesser extent, from peonidin conjugated with p-hydroxybenzoic acid, ferulic acid, or caffeic acid

(Kim et al., 2012). Although distinct from Ipomoea batatas, coloured-flesh potatoes similarly accumulate high levels of acylated anthocyanins, which contribute to pigment stability and potential health benefits.

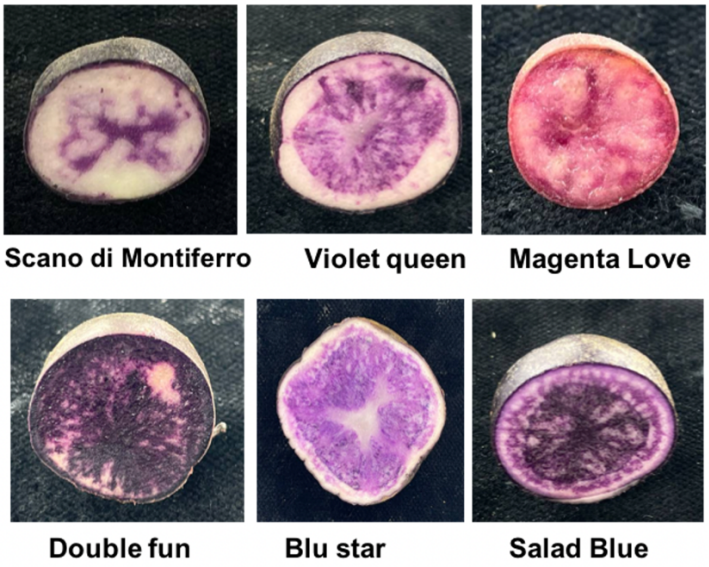

Figure 6. Different colored-flesh potato (*Solanum tuberosum* L.) cultivars characterized by a high anthocyanin content. The cultivars Scano di Montiferro, Violet Queen, Magenta Love, Double Fun, Blu Star, and Salad Blue display color variations resulting from differences in the composition and distribution of anthocyanins within the tubers. In particular, petunidins and malvidins are the predominant pigments in purple-fleshed varieties (Violet Queen, Blu Star, Salad Blue), whereas peonidins and pelargonidins prevail in red to pink varieties (Magenta Love).

In potato, the transcriptional control of anthocyanin biosynthesis largely conforms to the general MBW regulatory model described above but involves species-specific regulators. In particular, the R2R3-MYB transcription factor StAN1, in combination with bHLH and WD40 partners, forms an StAN1–bHLH–WD40 complex that activates late biosynthetic genes in a tissue-specific manner. Recent transcriptomic analyses confirmed the central role of the StAN1-bHLH-WD40 complex in tissue-specific induction, particularly in tuber eyes, where defense-related genes are co-expressed (Tang et al., 2023), suggesting a functional link between anthocyanin accumulation and stress-responsive pathways in potato. This dual role as a determinant of tuber pigmentation and as a potential mediator of stress adaptation makes potato anthocyanins an ideal trait for investigating the interplay between secondary metabolism, abiotic stress responses and, as will be discussed later, epigenetic regulation.

1.5 Epigenetic regulation via histone modifications and its role in metabolite adaptation to environmental stress

Epigenetics focuses on heritable changes in gene expression that occur without altering the underlying DNA sequences (Gallo-Franco et al., 2020). Epigenetic mechanisms encompass nucleosome positioning, DNA methylation, histone variants and modifications, and small RNAs, collectively defining the epigenome (Duan et al., 2018). Histone modifications warrant specific attention, as they involve chemical changes to the N-terminal tails of the histone proteins that play a key role in DNA packaging and gene expression regulation. They involve chemical changes that protrude from the nucleosome core, including acetylation, methylation, phosphorylation, ubiquitination, and glycosylation. These modifications establish specific epigenetic states that enable the genome to adapt to environmental changes (Zhang et al., 2008; Li et al., 2021). By altering chromatin accessibility and the recruitment of transcriptional regulators, histone marks can modulate not only genes directly involved in stress perception and signaling, but also those controlling primary and secondary metabolism, thereby contributing to metabolic plasticity under fluctuating environmental conditions. However, while certain aspects of histone modifications, such as methylation/demethylation and acetylation/deacetylation, have been studied, particularly for their influence on transcription by controlling the accessibility of transcription factors to DNA (Kumar et al., 2021; Zhang et al., 2018), their broader role in the epigenetic regulation of plant stress responses and metabolite-based adaptation to stress remains largely unexplored. A review by Liu et al. 2022 provides a comprehensive overview of the dynamics of DNA methylation and histone modifications in plant responses to abiotic stresses, offering insights into the epigenetic mechanisms involved. Building on this framework, the following subsections focus first on major classes of histone marks, methylation and demethylation (1.5.1), acetylation (1.5.2), histone variants and less well-known post-translational modifications (1.5.3) and then on how these modifications can function as a memory of stress-induced metabolic reprogramming, even across generations (1.5.4), with particular emphasis on their role in the epigenetic regulation of stress responses and metabolite adaptation in *S. tuberosum* (1.5.5).

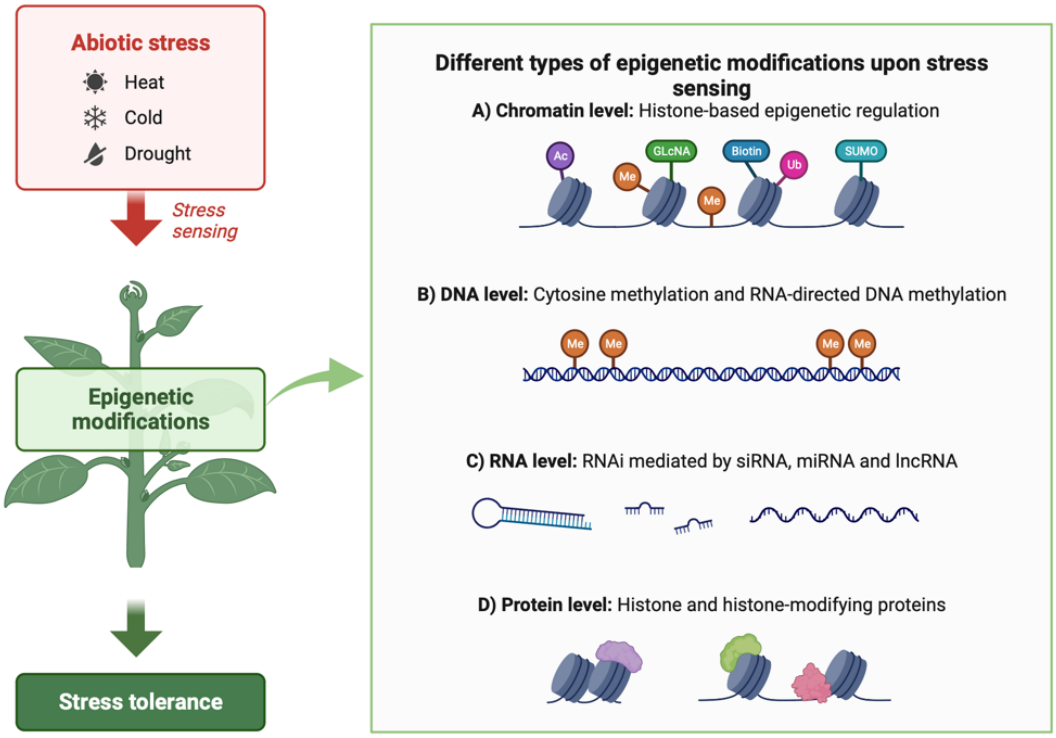

Figure 7. Epigenetic modifications in plants in response to abiotic stress. Schematic representation of different epigenetic mechanisms activated upon exposure to heat, cold, or drought stress. These include histone-based chromatin regulation (acetylation, methylation, ubiquitination, etc.), cytosine methylation and RNA-directed DNA methylation, RNA interference mediated by siRNA, miRNA, and lncRNA, as well as histone and histone-modifying proteins acting at the protein level. Image adapted from Singroha, G., & Sharma, P. (2019). Epigenetic modifications in plants under abiotic stress. In R. Meccariello (Ed.), Epigenetics. IntechOpen. https://doi.org/10.5772/intechopen.84455

1.5.1 Histone methylation and demethylation

Histone methylation and demethylation can either repress or activate gene expression, depending on the modification site. Methylation of lysine 4 on histone 3 (H3K4me3) and methylation of lysine 36 on histone 3 (H3K36/me3) promote transcription by opening chromatin, whereas methylation at lysine 9 (H3K9me2) and lysine 27 (H3K27me3) compacts chromatin and represses transcription (Kumar et al., 2021). Conversely, demethylation at these sites has opposite effects: removing methyl groups from H3K4me3 typically represses, while demethylating H3K9me2 or H3K27me3 activates gene expression by eliminating repressive methylation marks (Bhadouriya et al., 2020). These modifications are crucial for regulating developmental genes and for influencing

metabolite production in response to environmental stresses. For example, light exposure in apple (*Malus domestica*), leads to demethylation of histone H3K27me3, enhancing transcription of genes like *MdMYB10* and *MdMYB1* and promoting anthocyanin production (Bai et al., 2016). Similarly, the *JMJ25* gene, a histone H3K9 demethylase from *Populus trichocarpa*, is known to suppress anthocyanin biosynthesis. Its expression, induced under dark conditions, suggests a stress-response mechanism where plants conserve energy by reducing secondary metabolites like anthocyanins. ChIP-qPCR assays confirmed the role of *JMJ25* in dynamically demethylating H3K9me2 at the MYB182 chromatin (Fan et al., 2018). In *A. thaliana*, *JMJ15*, which encodes an H3K4 demethylase, enhances salt tolerance. Overexpression of this gene results in shortened plants and increased lignin in stems, which contributes to their salt resistance (Yung et al., 2021; Shen et al., 2014). Recently, research has increasingly focused on the divergence of chromomethylases and their role in plant adaptation to environmental stresses. For example, the divergence between *CMT2* and *CMT3*, two chromomethylases, has allowed them to specialize in distinct functions, contributing to DNA methylation regulation. Jiang et al. (2024) demonstrated that *CMT2* interacts with the modified histone H3K9me to guide CHH methylation in heterochromatic regions, contributing to transposon silencing and genome stability. Additionally, *CMT2* has an N-terminal region that regulates its stability under thermal stress, suggesting a possible role of these epigenetic mechanisms in plant responses to challenging environmental conditions.

1.5.2 Histone acetylation

Histone acetylation, such as that occurring at lysine residues H3K9 and H3K27, generally enhances gene expression by making chromatin more open and accessible to transcription factors. For example, salt stress induces acetylation of histone H3K9 in the promoter region of stress-responsive genes, contributing to their activation in plants like maize (*Zea mays*) and black cottonwood (*P. trichocarpa*) (Li et al., 2019; Li et al., 2023). In salt-stressed maize, plant response is accompanied by increased levels of global histone H3 acetylation at K9, which has been shown to control the increase in cell wall proteins such as expansins and the plasma membrane proton pump (Li et al., 2014). In contrast, deacetylation typically represses gene expression, highlighting a dynamic regulation of chromatin states in response to environmental conditions. Further studies have highlighted the role of histone acetylation in response to other stresses. For example, drought stress leads to a reduction in H3K9ac, which is associated with the

downregulation of genes involved in the synthesis of flavonoids and abscisic acid (ABA), both of which are crucial for plant stress responses. Treatment with the histone deacetylase inhibitor trichostatin A (TSA) attenuated these effects, enhancing gene expression linked to flavonoid and ABA synthesis, thus improving drought tolerance (Li et al., 2023). Additionally, UV-B irradiation increases H3K9 acetylation at flavonol metabolic pathway genes, enhancing their activity (Figure 8). This response can be modulated by treatment with the PAMP flg22 peptide, a bacterial immune response elicitor, which suppresses H3K9 acetylation and represses flavonol gene expression. Similar regulatory patterns have been observed in hemp (*Cannabis sativa*), where expression levels of Histone Acetyltransferase (HAT) genes significantly change in response to various abiotic stresses (e.g., cold and salt), impacting the synthesis of cannabinoids. Suppression of these HATs through treatment with the inhibitor PU139 has been shown to negatively impact cannabinoid production, highlighting the critical role of histone acetylation in managing plant responses to environmental stress (Cheng et al., 2023). A recent study by Yu et al. (2024) explores the role of lysine acetylation of the histone acetyltransferase adaptor protein ADA2 as a metabolic control mechanism that connects cellular acetyl-CoA levels to chromatin modifications in plants. Acetyl-CoA, a key metabolite in cellular metabolism, acts as a donor for acetyl groups in histone acetylation reactions. Research reveals that specific lysine residues on ADA2 are acetylated in response to fluctuations in acetyl-CoA levels, acting as a "sensor" to regulate histone acetyltransferase (HAT) activity. Indeed, when acetyl-CoA levels rise, ADA2 acetylation increases, boosting HAT activity and histone acetylation, which opens chromatin and activates stress-responsive genes. Conversely, lower acetyl-CoA levels reduce ADA2 acetylation, restricting HAT activity and chromatin accessibility.

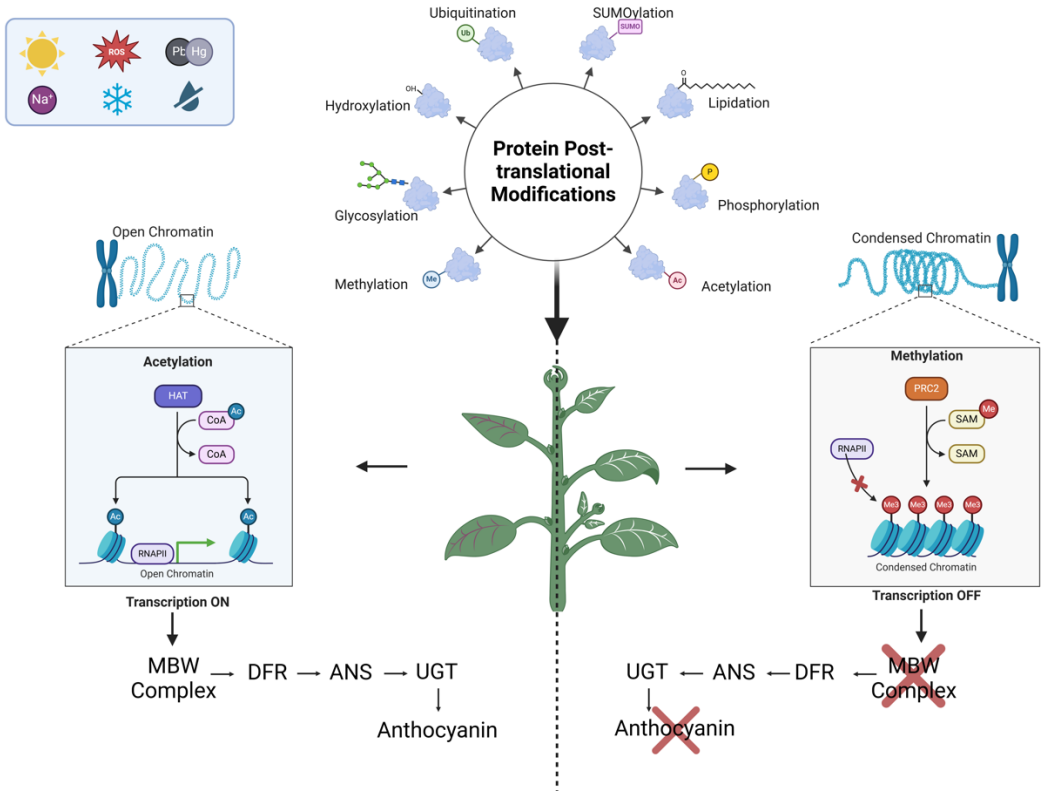

Figure 8. Abiotic factors trigger post-translational modifications (PTMs) in plants, influencing chromatin structure and gene expression. An example of anthocyanin biosynthesis is presented: on the left, acetylation (Ac) opens chromatin, allowing transcriptional activation of the MBW complex and downstream enzymes (*DFR*, *ANS*, *UGT*), leading to anthocyanin production. On the right, methylation (Me) condenses chromatin, inhibiting transcription of the same biosynthetic pathway, preventing anthocyanin accumulation. This highlights how PTMs, such as acetylation and methylation, regulate chromatin dynamics in response to environmental stimuli. Cimmino et al 2025 Created in BioRender: **https://BioRender.com/k98j551** (accessed on 2 December 2024).

1.5.3 Histone variants and less well-known PTMs

Recent studies have revealed an additional layer of complexity: histone variants such as H2A.Z can interact with histone modifications to further modulate gene activity. H2A.Z is a variant of the histone H2A, evolutionarily conserved, that integrates into nucleosomes and plays key roles in gene regulation and chromatin stability. The H2A.Z variants differ from canonical H2A at many positions throughout the primary sequence, with the main differences observed in the carboxy-terminal α-helix included in the docking domain. Specifically, in wild-type Arabidopsis plants SW1, the expression of anthocyanin biosynthesis genes is inhibited through the incorporation of H2A.Z into the nucleosomes of these genes. This incorporation hinders the accumulation of H3K4me3, a mark

associated with active transcription, and ultimately suppresses the expression of anthocyanin biosynthesis genes (Cai et al., 2019). Similarly, in mutants with defects in H2A.Z deposition (arp6/pie1/hta9 hta11), an elevation in H3K4me3 levels is observed. This increase is linked to enhanced expression of anthocyanin biosynthesis genes, causing accumulation of such pigments in these mutants (Cai et al., 2019). These findings underscore the intricate interplay between histone variants and modifications in regulating key metabolic pathways, suggesting that answers to these and related questions may clarify the mechanisms underlying metabolic regulation in plants.

Other less well-known histone modifications involve the covalent addition of molecules, such as phosphate groups, ubiquitin, and SUMO, which can influence gene expression and stress responses depending on the histone modifications site. For example, phosphorylation of histone H3 at Serine 10 (H3S10) is associated with transcriptional activation, whereas other phosphorylation events on histone H3 (e.g., H3S28ph and H3T3ph) can show context-dependent effects on chromatin state in plants, including during stress responses and cell-cycle transitions. Ubiquitination of histone lysine residues can act as a marker for both activation (e.g., H2Bub with H3K4me3) and repression (e.g., H2Aub with H3K27me3). In upland cotton (*Gossypium hirsutum*), ubiquitination has also been linked to the regulation of organ size (Wang et al., 2022). SUMOylation, in contrast, plays a role in responses to environmental stresses, as shown by the increased SUMO signals associated with chromatin during heat stress (Han et al., 2021).

Small RNAs (such as microRNAs, siRNAs, and phased siRNA (phasiRNA/tasiRNA) play a crucial role in modulating post-translational modifications (PTMs) and epigenetic regulation. These RNAs not only act as intermediaries in gene expression regulation but also closely interact with proteins involved in RNA methylation, such as plant m^{6}A regulators (e.g., the Arabidopsis writer components MTA/MTB/FIP37 and AlkB-family demethylases such as ALKBH9B/ALKBH10B, with m^{6}A “readers” including ECT proteins), serving as “readers”, “writers”, and “erasers”. For instance, these proteins are subjected to phosphorylation and ubiquitination influencing their catalytic activity, stability, and subcellular localization and, consequently, the modifications on methylated RNAs. Small-RNA pathways can intersect with RNA-modification and chromatin regulators (directly or indirectly), contributing to dynamic regulatory circuits that tune gene expression during stress acclimation. For example, stress-activated signaling (e.g., via kinases and ubiquitin ligases) may modulate the assembly/activity of the plant m^{6}A

writer complex (MTA/MTB/FIP37) and thereby reshape m^{6}A patterns on specific transcripts, with potential impacts on plant developmental programs (e.g., meristem activity) and abiotic-stress responses (Chen et al., 2023). Despite advancements in understanding these mechanisms, the role of these PTMs in metabolite biosynthesis under stress remains unclear, highlighting the need for further studies to elucidate their involvement in transcriptional control during plant adaptation to abiotic stresses. It would be interesting to further investigate how these modifications contribute to long-term gene expression changes in response to various environmental stresses. Understanding this could clarify how plants utilize epigenetic mechanisms for adaptive resilience.

1.5.4 Histone modifications serve as a memory for metabolic adaptation, even across generations

The retention of molecular experiences, often referred to as "stress priming", allows plants to recall past stress episodes and utilize the stored information for more efficient adaptation to new conditions (Turgut-Kara et al., 2020; Ling et al., 2018). The term "priming" has gained prominence, signifying that mild exposure to stress can activate plant stress responses, enabling faster and stronger reactions upon subsequent stress occurrences. This adaptive mechanism involves the establishment of an epigenetic memory that plays a crucial role in helping plants overcome future stress challenges (Ding et al., 2012; Ling et al., 2018; Gully et al., 2019). The duration of memory varies depending on the type of environmental exposure. Cold memory induced by vernalization can last until reproductive growth, while most stress-induced memories will be short-term if the stress does not recur. These signals not only modulate plant response to the surrounding environment but can also be maintained during plant development or leave an inheritable epigenetic imprint across generations. Indeed, memory can be divided into two main categories: somatic memory and transgenerational memory. The former involves epigenetic changes in an organism that occur in response to environmental stress. Evidence shows that PTMs play a key role in regulating somatic stress memory, particularly in relation to primary and secondary metabolism. For example, in meadow foxtail (*Alopecurus pratensis*), plants subjected to repeated waterlogging and drought stress demonstrated enhanced recovery. Indeed, they displayed higher levels of Rubisco content, antioxidative enzymes (POX, SOD), and chlorophyll b, all indicators of long-term drought stress memory (Lukic et al., 2020). These responses are tightly regulated by histone modifications. Under cold stress, Arabidopsis plants showed reduced H3K27me3

deposition at specific cold-responsive genes such as *COR15A* and *GALACTINOL SYNTHASE 3* (*GOLS3*), which regulate the biosynthesis of raffinose oligosaccharides (RFOs). This reduction persisted even after the stress, suggesting that H3K27me3 may function as an epigenetic marker for stress memory (Kwon et al., 2009). Similarly, drought memory in rice was associated with increased H3K4me3 levels observed at key genes involved in proline biosynthesis, crucial for stress tolerance (Ding et al., 2012). This indicates that histone modification is involved in plants' ability to "remember" and respond more effectively to future drought events during their life cycle. Salt stress also induces changes in the distribution of H3K27me3 marks, influencing somatic stress memory. This alteration allows the release of previously repressed genes, such as HKT1, enabling them to respond more effectively and rapidly to future salt stress episodes (Sani et al., 2013). Additionally, the maintenance of elevated levels of H3K4me3 at the locus of Δ1-pyrroline-5-carboxylate synthetase 1 (P5CS1), a key enzyme involved in proline synthesis during recovery after salt stress, further reinforces the role of histone modifications in establishing salt stress memory and ensuring better recovery from such conditions (Feng et al., 2016). Transgenerational memory, on the other hand, involves the transmission of epigenetic modifications across generations. It is a less well-studied and more elusive phenomenon compared to somatic memory. However, its comprehension and interpretation are less explored. Lukić et al. (2022) demonstrated that maternal stress experiences, such as drought or waterlogging, can precondition offspring to respond better to similar stress, with higher biomass and phenolic compounds production. This suggests a possible link between epigenetic marks, including PTMs, and metabolite regulation in stress response. Although research shows that histone modifications can influence secondary metabolite biosynthesis under stress (Table 1), it remains uncertain whether this regulation forms a memory of prior biosynthetic states. Further investigation into how PTMs store information about metabolite production could unlock opportunities to enhance the production of important compounds in agriculture and medicine.

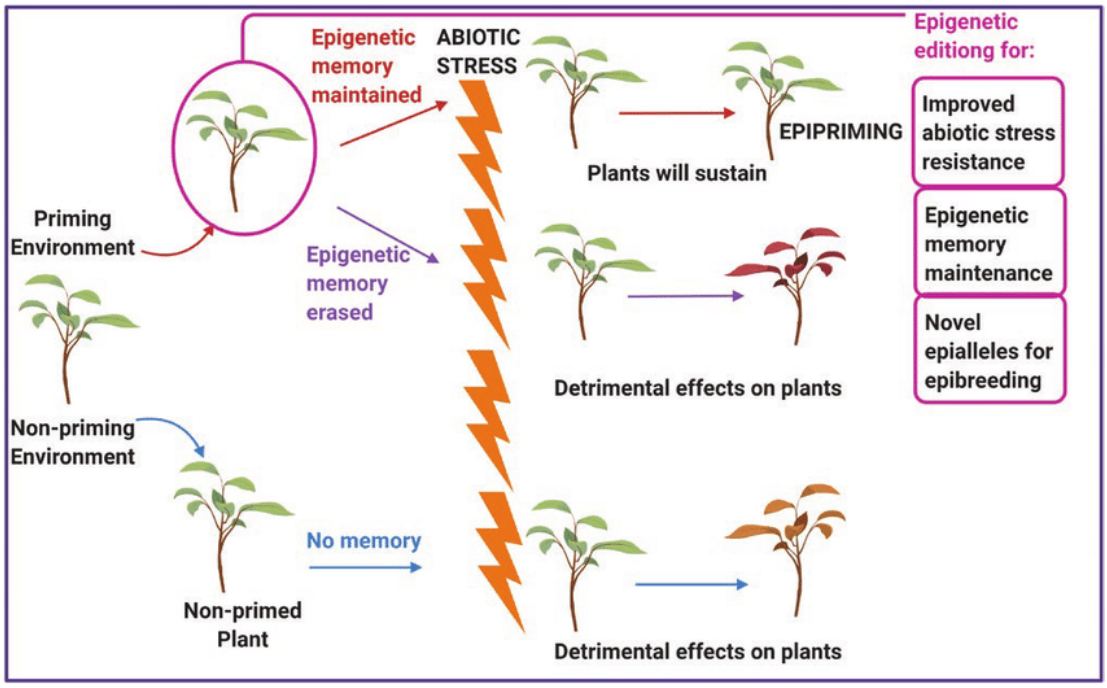

Figure 9. Epi-priming for crop resilience towards abiotic stresses. Retained epigenetic memory helps in plant sustenance under subsequent stress, making the plant epigenetically primed. This figure was created with Biorender.com. Downloaded from https://academic.oup.com/jxb/advance-article/doi/10.1093/jxb/erab337/6327780 by Indian Agricultural Research Institute Library (IARI) user on 24 October 2021

Table 1. Summary of histone modifications and PTMs in different plant species and their influence on secondary metabolite production under abiotic stress conditions.

Histone modification	Plant species	Secondary metabolites	Histone mark	Abiotic stressor	Reference
Demethylation	*M. domestica*	Lignin	H3K4me3	Salt	Bai et al., 2016
Acetylation	*A. thaliana*	Flavonol	H3K9ac	UV-B	Gully et al., 2019
Demethylation	*Populus spp.*	Anthocyanin	H3K9me2	Dark	Kumar et al., 2023
Methylation	*A. thaliana*	Anthocyanin	H3K4me3	-	Li et al., 2014
Acetylation	*Cannabis sativa*	Cannabinoid	H3K9ac	Cold/salt	Jiang et al., 2024
Deacetylation	*Hippophae rhamnoides*	Flavonoids and (ABA)	H3K9ac deacetylated	Drought	Shen et al., 2014

1.5.5 Epigenetic regulation of stress responses in Solanum tuberosum

In the *Solanum* genus, a growing number of studies highlight the central role of epigenetics in regulating stress responses. In potato, for example, the inhibition of DNA methylation through zebularine accelerates tuber induction by acting on key genes involved in photoperiod and hormone pathways (Xu et al., 2021). Similarly, methylation of the promoter of the *OMT30376* gene, involved in anthocyanin modification, has been identified as the cause of color differences between purple and red tubers (Liu et al., 2022). This example demonstrates how reversible epigenetic modifications can modulate complex physiological processes that are highly dependent on environmental conditions and can directly influence the biosynthesis of secondary metabolites, linking epigenetic variation to visible phenotypes.

Additional studies show that abiotic stress, particularly cold, induces bivalent histone modifications (H3K4me3 and H3K27me3) on genes activated during stress exposure (Zhou et al., 2024). The simultaneous presence of activating and repressive marks suggests a flexible regulatory mechanism that enables genes to respond rapidly to environmental changes. Recent evidence also indicates that drought priming can cause stable changes in the methylome of potato clones, promoting faster transcriptional responses and improved performance during later growth stages (Hernández et al., 2023), reflecting a form of stress memory based on epigenetic modulation. In parallel, small RNA atlases generated under salt stress conditions have identified miR408 and miR169a as key regulatory nodes involved in lignification and ion homeostasis, confirming the role of small RNAs as crucial components of the epigenetic response to stress (Wang et al., 2022).

Recent work further demonstrates that in potato epigenetic regulation under stress extends beyond DNA methylation and small RNAs, involving dynamic regulation of DNA methyltransferases and demethylases in response to temperature, and likely integration of chromatin-remodeling and histone-modification pathways. For instance, under high-temperature stress, cultivars differing in heat tolerance show differential expression of CMT and DRM DNA methyltransferases and demethylases; the heat-tolerant genotype shows upregulation of demethylases and RdDM pathway components that correlate with expression changes of tuberization regulators (e.g., *StSP6A*, *StBEL5*) (Dutta et al., 2022). This suggests that active DNA demethylation and small-RNA–directed methylation pathways may underlie acclimation of potato to heat stress via epigenetic reprogramming of developmental regulators.

Moreover, genome-wide analyses across plants indicate that epigenetic mechanisms (DNA methylation dynamics, histone modifications, chromatin remodeling, and small RNAs) act as an integrated network modulating gene expression under diverse abiotic stresses (drought, salinity, heat), and recent reviews propose that similar mechanisms operate in potato and other major crops (Abdulraheem et al., 2024; Yang et al., 2025). Therefore, epigenetic regulation provides a versatile, multilayered and reversible interface between environmental signals and gene expression, which could be exploited to improve stress resilience in potato. The overall picture emerging from this body of evidence is that the link between epigenetics, stress regulation and secondary metabolism is strong, suggesting that epigenetic modulation represents a promising strategy to improve stress resilience in potato and other Solanaceae crops (Chen et al., 2025).

1.6 Aim of the thesis

Understanding how plants respond and adapt to abiotic stress is essential for improving crop resilience and productivity in the context of increasingly rapid climate change. In this scenario, *S. tuberosum* L. represents not only a globally important food crop but also a model species for investigating metabolic and molecular stress responses. Among its wild relatives, *S. commersonii* is of particular interest because it exhibits notable tolerance to several abiotic stresses and displays distinctive morpho-physiological traits that make it a valuable comparative system for studying mechanisms of environmental adaptation. Anthocyanins, glycosylated and water-soluble flavonoids that accumulate mainly in the vacuoles, play a key role in protecting plant cells thanks to their antioxidant capacity, osmoprotective functions and involvement in signaling pathways. Alongside these metabolic components, higher regulatory layers such as regulatory RNAs and histone modifications also contribute significantly to shaping gene expression patterns during stress responses, dynamically influencing chromatin accessibility and transcriptional activity. To reconstruct how anthocyanin metabolism and multiple levels of gene regulation contribute to abiotic stress responses, this work follows a progressive and integrated approach across three experimental chapters, each focused on a different stress and combination of Solanum species and methodological depth (Figure 10).

- In Chapter II, the study focused exclusively on cultivated *S. tuberosum* cultivars and on drought stress to evaluate their physiological responses to drought and to

understand how intraspecific genetic variability affects tolerance to water deficit. Four cultivars differing in tuber pigmentation and anthocyanin content (two yellow-fleshed, non-pigmented and two anthocyanin-rich genotypes) are compared under controlled water deficit and recovery. The aim here is to (i) evaluate how intraspecific genetic variability in anthocyanin accumulation influences drought responses at the whole-plant level, (ii) characterize associated changes in primary and secondary metabolism, and (iii) assess the expression of key genes of the phenylpropanoid/anthocyanin pathway and stress-related markers. This chapter, therefore, provides a first, "classical" layer of information on the role of anthocyanin-rich genotypes in *S. tuberosum* drought tolerance, based on detailed physiology and targeted gene expression rather than genome-wide approaches.

- Building on these results, Chapter III broadens the perspective to salinity stress and interspecific comparison and introduces next-generation sequencing to explore regulatory networks. Here, the salt-susceptible *S. tuberosum* cv. Désirée is compared with the wild salt-tolerant *S. commersonii* clone 1T under prolonged salt stress. The objective is to (i) dissect contrasting salt-stress phenotypes through gas-exchange measurements, pigment quantification and biochemical stress biomarkers, and (ii) determine how these physiological differences are underpinned by transcriptomic and epigenetic reprogramming. RNA sequencing (RNA-seq) is used to identify differentially expressed genes and enriched pathways, with particular attention to anthocyanin biosynthesis, phenylpropanoid metabolism and major stress-responsive processes. In parallel, small RNA sequencing is employed to characterize regulatory RNAs (microRNAs and siRNAs) potentially involved in post-transcriptional control, while ChIP-seq and ChIP-qPCR are used to map and validate key histone modifications associated with stress-responsive and anthocyanin-related genes. Together, these datasets provide a multilayered view of how metabolic, transcriptional and epigenetic regulation jointly contribute to the superior salt resilience of *S. commersonii* compared with cultivated potato.

- Finally, Chapter IV evaluated the role of priming in stress memory under an additional abiotic stress that is particularly harmful for crop productivity, heat stress. The analysis was extended beyond cultivated potato to another Solanaceae species, *S. lycopersicum*. The experiment was originally conceived as a

comparative analysis including *S. tuberosum* cv. Désirée; however, technical limitations in the potato datasets (uneven survival and suboptimal RNA quality) prevented a robust genome-wide analysis. Consequently, this chapter presents and discusses in detail the results obtained in tomato, which becomes the model system for investigating heat-stress priming. The heat-stress priming experiment presented in this chapter was carried out during a six-month research stay in the laboratory of Prof. Moussa Benhamed (Chromatin and Development, Université Paris-Saclay), where I established and performed the priming protocol in both potato and tomato. Priming-induced transcriptional changes were investigated in order to understand how stress memory influences gene expression regulation and to provide insights useful for the genetic improvement of plants, ultimately aimed at enhancing resistance to abiotic stresses. In particular, RNA-seq and network-based analyses are used to identify transcriptional programs and regulatory modules associated with priming-induced stress memory, including those related to hormone signaling, heat-shock responses and secondary metabolism.

Overall, this thesis presents a comprehensive and integrated framework for understanding plant responses and adaptation to abiotic stress within the genus *Solanum* (Figure 10) By moving from drought in cultivated potato (Chapter II, targeted physiological and molecular analyses), to salinity in cultivated vs. wild *Solanum* with combined transcriptomic and epigenetic profiling (Chapter III), and finally to heat-stress priming in tomato using genome-wide expression analyses (Chapter IV), this work progressively incorporates additional biological complexity and methodological resolution. In this perspective, flavonoid metabolism, wild-species-derived traits and stress priming emerge as fundamental resources for the development of crops with enhanced resilience to environmental stresses, while regulatory RNAs and histone modifications provide key mechanistic links between environmental cues, gene expression and metabolic adaptation.

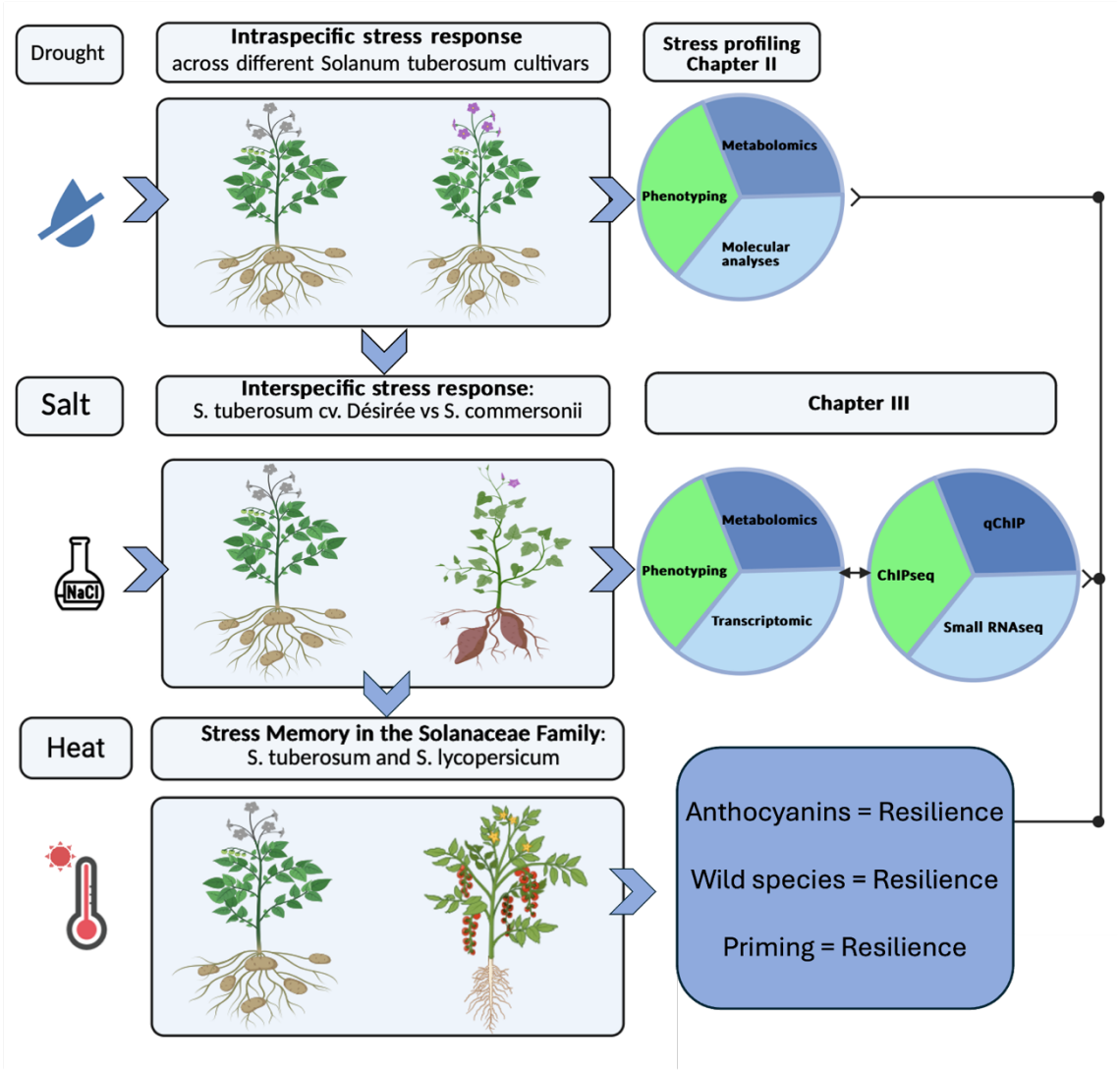

Figure 10. Schematic representation of the multi-layered approach adopted in this thesis. Intraspecific stress profiling of different *S. tuberosum* cultivars under drought (Chapter II) and interspecific comparison between *S. tuberosum* cv. Désirée and the wild species *S. commersonii* under salt stress (Chapter III) are combined with epigenomic analyses, including small RNA sequencing, ChIP-seq and ChIP-qPCR, within the salt-stress experiment (Chapter III). In addition, stress memory responses were investigated in the Solanaceae crop *S. lycopersicum* under heat stress (Chapter IV) to evaluate how transcriptional and epigenetic priming contribute to enhanced stress resilience. This integrated strategy was used to investigate how phenotypic traits, gene expression, and epigenetic regulation contribute to stress resilience, with particular focus on the role of anthocyanins, wild species-derived adaptive mechanisms and stress memory.

References

Abdulraheem, M. I., Xiong, Y., Moshood, A. Y., Cadenas-Pliego, G., Zhang, H., & Hu, J. (2024). Mechanisms of plant epigenetic regulation in response to plant stress: recent discoveries and implications. *Plants*, *13*(2), 163.

Abe, H., Yamaguchi-Shinozaki, K., Urao, T., Iwasaki, T., Hosokawa, D., & Shinozaki, K. (1997). Role of Arabidopsis MYC and MYB homologs in drought- and abscisic acid–regulated gene expression. *The Plant Cell, 9*(10), 1859–1868.

Almeida, P., Katschnig, D., & De Boer, A. H. (2013). HKT transporters—state of the art. *International journal of molecular sciences*, *14*(10), 20359-20385.

Andre, C. M., Schafleitner, R., Legay, S., Lefevre, I., Aliaga, C. A., Nomberto, G., Hoffmann, L., Hausman, J. F., Larondelle, Y., & Evers, D. (2007). Antioxidant profiling of native Andean potato tubers (Solanum tuberosum L.) reveals cultivars with high levels of β-carotene, α-tocopherol, chlorogenic acid, and petanin. Journal of Agricultural and Food Chemistry, 55(26), 10839–10849.

Arıkan, B., Çakır, Ö., & Kara, N. T. (2025). Epigenetic and transcriptomic features of stress memory during salt-stress priming in arabidopsis. *Plant Science*, 112775.

Aversano, R., Contaldi, F., Ercolano, M. R., Grosso, V., Iorizzo, M., Tatino, F., ... & Carputo, D. (2015). The Solanum commersonii genome sequence provides insights into adaptation to stress conditions and genome evolution of wild potato relatives. *The Plant Cell*, *27*(4), 954-968.

Aversano, R., D'Amelia, V., Esposito, S., Garramone, R., Villano, C., & Carputo, D. (2021). *The Solanum commersonii genome sequence*. In D. Carputo, R. Aversano, & M. R. Ercolano (Eds.), The wild Solanums genomes (pp. 167–180). Springer Nature.

Bai, S., Tuan, P.A., Saito, T., Honda, C., Hatsuyama, Y., Ito, A., & Moriguchi, T. (2016). Epigenetic regulation of MdMYB1 is associated with paper bagging-induced red pigmentation of apples. Planta, 244, 573–586.

Barbaś, P. I. O. T. R., Noaema, A. H., & Sawicka, B. A. R. B. A. R. A. (2023). Potato (Solanum tuberosum L.) as a rich source of nutrients and bioactive compounds: a review. *Journal of Cell and Tissue Research*, *23*(2), 7337-7355.

Batool, T., Arif, M. S., Yasmeen, T., Riaz, M., Shahzad, S. M., & Asif, M. (2020). Plant growth promoting rhizobacteria alleviates drought stress in potato in response to suppressive oxidative stress and antioxidant enzyme activities. Scientific Reports, 10, 16975.

Bhadouriya, S.L., Mehrotra, S., Basantani, M.K., Loake, G.J., & Mehrotra, R. (2020). Role of chromatin architecture in plant stress responses: An update. Frontiers in Plant Science, 11, 603380.

Bhargava, S., & Sawant, K. (2013). Drought stress adaptation: Metabolic adjustment and regulation of gene expression. *Plant Breeding, 132*(1), 21–32.

Blumwald, E. (2000). Sodium transport and salt tolerance in plants. *Current opinion in cell biology*, *12*(4), 431-434.Boyer, J. S. (1982). Plant productivity and environment. Science, 218(4571), 443–448.

Bradshaw, J. E., & Bonierbale, M. (2010). Potatoes. In Root and Tuber Crops (pp. 1–52). Springer, New York.

Cai, H., Zhang, M., Chai, M., He, Q., Huang, X., Zhao, L., & Qin, Y. (2019). Epigenetic regulation of anthocyanin biosynthesis by an antagonistic interaction between H2A.Z and H3K4me3. New Phytologist, 221, 295–308.

Carputo, D., Barone, A., Cardi, T., Sebastiano, A., Frusciante, L., & Peloquin, S. J. (1997). Endosperm balance number manipulation for direct in vivo germplasm introgression to potato from a sexually isolated relative (*Solanum commersonii*Dun.). *Proceedings of the National Academy of Sciences of the United States of America*, **94**(22), 12013–12017. https://doi.org/10.1073/pnas.94.22.12013

Carputo, D., Frusciante, L., & Peloquin, S. J. (2003). The role of 2n gametes and endosperm balance number in the origin and evolution of polyploids in the tuber-bearing Solanums. Genetics, 163(1), 287–294.

Chalker-Scott, L. (1999). Environmental significance of anthocyanins in plant stress responses. Photochemistry and Photobiology, 70(1), 1–9.

Charng, Y.-Y., Liu, H.-C., Liu, N.-Y., Hsu, F.-C., & Ko, S.-S. (2007). *A heat-inducible transcription factor, HsfA2, is required for extension of acquired thermotolerance in Arabidopsis.* Plant Physiology, 143, 251–262.

Chaves, M. M., Maroco, J. P., & Pereira, J. S. (2003). Understanding plant responses to drought—from genes to the whole plant. Functional Plant Biology, 30(3), 239–264.

Chen, H. H., & Li, P. H. (1980). Biochemical changes in tuber-bearing Solanum species in relation to frost hardiness during cold acclimation. *Plant Physiology*, *66*(3), 414-421.

Chen, X., Ding, Y., Yang, Y., Song, C., & Gong, Z. (2021). Protein kinases in plant responses to drought, salt, and cold stress. *Journal of Integrative Plant Biology, 63*(1), 53–78.

Chen, Y., Jiang, Z., Yang, Y., Zhang, C., Liu, H., & Wan, J. (2023). The functions and mechanisms of post-translational modification in protein regulators of RNA methylation: Current status and future perspectives. International Journal of Biological Macromolecules, 253, 126773.

Cheng, Y., Ning, K., Chen, Y., Hou, C., Yu, H., & Guo, X. (2023). Identification of histone acetyltransferase genes responsible for cannabinoid synthesis in hemp. Chinese Medicine, 18, 16.

Cutler, S. R., Rodriguez, P. L., Finkelstein, R. R., & Abrams, S. R. (2010). Abscisic acid: Emergence of a core signaling network. Annual Review of Plant Biology, 61, 651–679.

Deblonde, P. M. K., & Ledent, J.-F. (2001). Effects of moderate drought conditions on green leaf number, stem height, leaf length and tuber yield of potato cultivars. European Journal of Agronomy, 14(1), 31–41.

Deinlein, U., Stephan, A. B., Horie, T., Luo, W., Xu, G., & Schroeder, J. I. (2014). Plant salt-tolerance mechanisms. *Trends in plant science*, *19*(6), 371-379.

Ding, Y., Fromm, M., & Avramova, Z. (2012). Multiple exposures to drought 'train' transcriptional responses in Arabidopsis. Nature Communications, 3, 740.

Duan, C.G., Zhu, J.K., & Cao, X. (2018). Retrospective and perspective of plant epigenetics in China. Journal of Genetics and Genomics, 45, 621–638.

Dutta, M., Raturi, V., Gahlaut, V., Kumar, A., Sharma, P., Verma, V., ... & Zinta, G. (2022). The interplay of DNA methyltransferases and demethylases with tuberization genes in potato (Solanum tuberosum L.) genotypes under high temperature. *Frontiers in Plant Science*, *13*, 933740.

Efimova, M. V., et al. (2018). Physiological mechanisms of Solanum tuberosum L. plants' tolerance to chloride salinity. Russian Journal of Plant Physiology, 65(3), 394–403.

Eiasu, B. K., Soundy, P., & Hammes, P. S. (2007). Response of potato (Solanum tuberosum) tuber yield components to gel-polymer soil amendments and irrigation regimes. New Zealand Journal of Crop and Horticultural Science, 35(1), 25–31.

Ekanayake, I. J., De Datta, S. K., & Steponkus, P. L. (1989). Spikelet sterility and flowering response of rice to water stress at anthesis. Annals of Botany, 63(2), 257–264.

Evers, D., Lefevre, I., Legay, S., Lamoureux, D., Hausman, J. F., Rosales, R. O. G., Marca, L. R. T., Hoffmann, L., Bonierbale, M., & Schafleitner, R. (2010). Identification of drought-responsive compounds in potato through a combined transcriptomic and targeted metabolite approach. *Journal of Experimental Botany, 61*(10), 2327–2343.

Fahad, S., et al. (2017). Crop production under drought and heat stress: Plant responses and management options. Frontiers in Plant Science, 8, 1147.

Fan, D., Wang, X., Tang, X., Ye, X., Ren, S., Wang, D., & Luo, K. (2018). Histone H3K9 demethylase JMJ25 epigenetically modulates anthocyanin biosynthesis in poplar. The Plant Journal, 96, 1121–1136.

Feng, X.J., Li, J.R., Qi, S.L., Lin, Q.F., Jin, J.B., & Hua, X.J. (2016). Light affects salt stress-induced transcriptional memory of P5CS1 in Arabidopsis. Proceedings of the National Academy of Sciences USA, 113, E8335–E8343.

Gallo-Franco, J. J., Sosa, C. C., Ghneim-Herrera, T., & Quimbaya, M. (2020). Epigenetic control of plant response to heavy metal stress: A new view on aluminum tolerance. Frontiers in Plant Science, 11, 602625.

Gebhardt, C. (2016). The historical role of species from the Solanaceae plant family in genetic research. Theoretical and Applied Genetics, 129(12), 2281–2294.

Gholizadeh, F., et al. (2025). Enhancing wheat resilience to salt stress through an integrative nanotechnology approach with chitosan proline and chitosan glycine. Scientific Reports, 15(1), 11126.

Golldack, D., Li, C., Mohan, H., & Probst, N. (2014). Tolerance to drought and salt stress in plants: unraveling the signaling networks. *Frontiers in plant science*, *5*, 151.

González, M., Galván, G. A., Siri, M. I., Borges, A., & Vilaró, F. (2013). Resistance to bacterial wilt of potato in *Solanum commersonii*. *Agrociencia Uruguay*, **17**(1), 45–54.

Goswami, G., Nath, U. K., Park, J.-I., Hossain, M. R., Biswas, M. K., Kim, H.-T., Kim, H. R., & Nou, I.-S. (2018). Transcriptional regulation of anthocyanin biosynthesis in a high-anthocyanin resynthesized Brassica napus cultivar. Journal of Biological Research, 25, 19.

Gould, K. S. (2004). Nature's Swiss army knife: The diverse protective roles of anthocyanins in leaves. BioMed Research International, 2004(5), 314–320.

Han, D., Chen, C., Xia, S., Liu, J., Shu, J., Nguyen, V., Lai, J., Cui, Y., & Yang, C. (2021). Chromatin-associated SUMOylation controls the transcriptional switch between plant development and heat stress responses. Plant Communications, 2, 100091.

Hancock, R.D., Morris, W.L., Ducreux, L.J., Morris, J.A., Usman, M., Verrall, S.R., Fuller, J., Simpson, C.G., Zhang, R., & Hedley, P.E. (2014). Physiological, biochemical and molecular responses of the potato (Solanum tuberosum L.) plant to moderately elevated temperature. Plant, Cell & Environment, 37, 439–450.

Hauser, F., & Horie, T. (2010). A conserved primary salt tolerance mechanism mediated by HKT transporters: a mechanism for sodium exclusion and maintenance of high K+/Na+ ratio in leaves during salinity stress. *Plant, cell & environment*, *33*(4), 552-565.

Hawkes, J. G. (1990). *The potato: Evolution, biodiversity and genetic resources*. Belhaven Press.

Hmida-Sayari, A., et al. (2005). Overexpression of Δ1-pyrroline-5-carboxylate synthetase increases proline production and confers salt tolerance in transgenic potato plants. Plant Science, 169(4), 746–752.

Hu, H., Dai, M., Yao, J., Xiao, B., Li, X., Zhang, Q., & Xiong, L. (2006). Overexpressing a NAM, ATAF, and CUC (NAC) transcription factor enhances drought resistance and salt tolerance in rice. *Proceedings of the National Academy of Sciences, 103*(35), 12987–12992.

Huimin Liu, Zijin Liu, Yu Wu, Lamei Zheng, & Genfa Zhang. (2021). Regulatory mechanisms of anthocyanin biosynthesis in apple and pear. International Journal of Molecular Sciences, 22, 8441.

International Panel on Climate Change. (2012). Summary for policymakers. In Managing the Risks of Extreme Events and Disasters to Advance Climate Change Adaptation (Field, C.B. et al., Eds.). Cambridge University Press, Cambridge, UK, p. 13.

Ismail, A. M., & Horie, T. (2017). Genomics, physiology, and molecular breeding approaches for improving salt tolerance. *Annual review of plant biology*, *68*, 405-434.

Jansky, S. H. (2006). Overcoming hybridization barriers in potato. *Plant Breeding*, **125**(1), 1–12. https://doi.org/10.1111/j.1439-0523.2006.01178.x

Jefferies, R. A., & MacKerron, D. K. L. (1993). Responses of potato genotypes to drought. II. Leaf area index, growth and yield. Annals of Applied Biology, 122(1), 105–112.

Kégbé, A. M., Salako, K. V., Enagnon Lokonon, B., Mensah, S., Noba, K., & Assogbadjo, A. E. (2025). A comparative study of morpho-physiological responses of wild and cultivated Solanum species to water stress: the case of S. sisymbriifolium and S. macrocarpon. *Genetic Resources and Crop Evolution*, *72*(2), 2249-2263.

Kim, H. W., et al. (2012). Anthocyanin changes in the Korean purple-fleshed sweet potato, Shinzami, as affected by steaming and baking. Food Chemistry, 130(4), 966–972.

Koes, R., Verweij, W., & Quattrocchio, F. (2005). Flavonoids: A colorful model for the regulation and evolution of biochemical pathways. Trends in Plant Science, 10(5), 236–242.

Kumar, S., Asrey, R. A. M., & Mandal, G. (2007). Effect of differential irrigation regimes on potato (Solanum tuberosum) yield and post-harvest attributes. Indian Journal of Agricultural Sciences, 77, 366–368.

Lachman, J., et al. (2009). Major factors influencing antioxidant contents and antioxidant activity in grapes and wines. International Journal of Wine Research, 1, 101–121.

Levy, D., & Veilleux, R.E. (2007). Adaptation of potato to high temperatures and salinity: A review. American Journal of Potato Research, 84, 487–506.

Li, D., Yang, Y., Li, Y.P., Zhu, X.H., & Li, Z.Q. (2021). Epigenetic regulation of gene expression in response to environmental exposures: From bench to model. Science of the Total Environment, 776, 145998.

Li, H., Yan, S., Zhao, L., Tan, J., Zhang, Q., Gao, F., Wang, P., Hou, H., & Li, L. (2014). Histone acetylation associated up-regulation of the cell wall related genes is involved in salt stress induced maize root swelling. BMC Plant Biology, 14, 105.

Li, J., et al. (2024). A novel Populus euphratica DUB gene, PeMINDY3, enhances drought and salt tolerance by promoting ROS scavenging. Environmental and Experimental Botany, 220, 105686.

Li, J., Wei, J., Song, Y., Chen, N., Ni, B., Zhang, J., & He, C. (2023). Histone H3K9 acetylation modulates gene expression of key enzymes in the flavonoid and abscisic acid pathways and enhances drought resistance of sea buckthorn. Physiologia Plantarum, 175, e13936.

Ling, Y., Serrano, N., Gao, G., Atia, M., Mokhtar, M., Woo, Y.H., Bazin, J., Veluchamy, A., Benhamed, M., & Crespi, M. (2018). Thermopriming triggers splicing memory in Arabidopsis. Journal of Experimental Botany, 69, 2659–2675.

Liu, H.-C., Liao, H.-T., & Charng, Y.-Y. (2011). *The role of class A1 heat shock factors (HSFA1s) in response to heat and other stresses in Arabidopsis.* Plant, Cell & Environment, 34, 738–751.

Liu, J., Sun, N., Liu, M., Liu, J., Du, B., Wang, X., & Qi, X. (2013). *An autoregulatory loop controlling Arabidopsis HsfA2 expression: role of heat shock–induced alternative splicing.* Plant Physiology, 162, 512–521.

Liu, Y., Wang, J., Liu, B., & Xu, Z.Y. (2022). Dynamic regulation of DNA methylation and histone modifications in response to abiotic stresses in plants. Journal of Integrative Plant Biology, 64, 2252–2274.

Lukić, N., Schurr, F.M., Trifkovic, T., Kukavica, B., & Walter, J. (2022). Transgenerational stress memory in plants is mediated by upregulation of the antioxidative system. SSRN Electronic Journal, 205, 105129.

Ma, Y., et al. (2021). Light-induced regulation pathway of anthocyanin biosynthesis in plants. International Journal of Molecular Sciences, 22(20), 11116.

Micheletto, S., Boland, R., & Huarte, M. (2000). Argentinian wild diploid *Solanum* species as sources of quantitative late blight resistance. *Theoretical and Applied Genetics*, **101**(5–6), 902–906. https://doi.org/10.1007/s001220051560

Mishra, S. K., Tripp, J., Winkelhaus, S., Tschiersch, B., Theres, K., Nover, L., & Scharf, K.-D. (2002). *In the complex family of heat stress transcription factors, HsfA1 has a unique role as master regulator of thermotolerance in tomato.*Genes & Development, 16, 1555–1567.

Molteberg, E.L. (2017). Influence of soil characteristics on skin finish of Norwegian potatoes. In Proceedings of the 20th Triennial EAPR Conference, Versaille, France, 9–14 July.

Motti, R. (2021). The Solanaceae Family: Botanical Features and Diversity. Springer International Publishing. https://doi.org/10.1007/978-3-030-30343-3_1

Muñiz García, M. N., Giammaria, V., Grandellis, C., Téllez-Iñón, M. T., Ulloa, R. M., & Capiati, D. A. (2012). Characterization of StABF1, a stress-responsive bZIP transcription factor from *Solanum tuberosum* L. that is phosphorylated by StCDPK2 in vitro. *Planta, 235*(4), 761–778.

Munns, R., & Tester, M. (2008). Mechanisms of salinity tolerance. Annual Review of Plant Biology, 59, 651–681.

Nakabayashi, R., et al. (2014). Enhancement of oxidative and drought tolerance in Arabidopsis by overaccumulation of antioxidant flavonoids. The Plant Journal, 77(3), 367–379.

Obidiegwu, J. E., et al. (2015). Coping with drought: Stress and adaptive responses in potato and perspectives for improvement. Frontiers in Plant Science, 6, 542.

Olmstead, R. G., Bohs, L., Migid, H. A., Santiago-Valentin, E., Garcia, V. F., & Collier, S. M. (2008). A molecular phylogeny of the Solanaceae. Taxon, 57(4), 1159–1181.

Sani, E., Herzyk, P., Perrella, G., Colot, V., & Amtmann, A. (2013). Hyperosmotic priming of Arabidopsis seedlings establishes a long-term somatic memory accompanied by specific changes of the epigenome. Genome Biology, 14, R59.

Saravia, D., et al. (2016). Yield and physiological response of potatoes indicate different strategies to cope with drought stress and nitrogen fertilization. American Journal of Potato Research, 93(3), 288–295.

Schafleitner, R., et al. (2009). The physiological responses of the potato (Solanum tuberosum) to drought stress are dependent on genotype and development stage. Field Crops Research, 114(1), 1–13.

Shen, Y., Conde, E.S.N., Audonnet, L., Servet, C., Wei, W., & Zhou, D.X. (2014). Over-expression of histone H3K4 demethylase gene JMJ15 enhances salt tolerance in Arabidopsis. Frontiers in Plant Science, 5, 290.

Singroha, G., Kumar, S., Gupta, O. P., Singh, G. P., & Sharma, P. (2022). Uncovering the epigenetic marks involved in mediating salt stress tolerance in plants. *Frontiers in Genetics*, *13*, 811732.

Szabolcs, I. (1994). Prospects of soil salinity for the 21st century. Agrokémia és Talajtan, 43(1–2), 5–24.

Tang, R., He, Y., & Fan, K. (2023). Recent advances in stability improvement of anthocyanins by efficient methods and its application in food intelligent packaging: A review. Food Bioscience, 56, 103164. https://doi.org/10.1016/j.fbio.2023.103164

Turgut-Kara, N., Arikan, B., & Celik, H. (2020). Epigenetic memory and priming in plants. Genetica, 148, 47–54.

Varga, I., & Djurovic, V. (2023). Trends of potato production and consumption in the world.

Villegas, D., et al. (2016). Exogenous application of pectin-derived oligosaccharides to grape berries modifies anthocyanin accumulation, composition and gene expression. Plant Physiology and Biochemistry, 104, 125–133.

Wang, P., Guo, K., Su, Q., Deng, J., Zhang, X., & Tu, L. (2022). Histone ubiquitination controls organ size in cotton (Gossypium hirsutum). The Plant Journal, 110, 1005–1020.

Winkel-Shirley, B. (2002). Biosynthesis of flavonoids and effects of stress. Current Opinion in Plant Biology, 5(3), 218–223.

Wolf, S., Marani, A., & Rudich, J. (1991). Effect of temperature on carbohydrate metabolism in potato plants. Journal of Experimental Botany, 42, 619–625.

Yang, X., Liu, J., Xu, J., Duan, S., Wang, Q., Li, G., & Jin, L. (2019). Transcriptome profiling reveals effects of drought stress on gene expression in diploid potato genotype P3-198. *International Journal of Molecular Sciences, 20*(4), 852.

Yoshida, T., Ohama, N., Nakajima, J., Kidokoro, S., Mizoi, J., Nakashima, K., … Yamaguchi-Shinozaki, K. (2011). *Arabidopsis HsfA1 transcription factors function as the main*

positive regulators in heat shock-responsive gene expression. Molecular Genetics and Genomics, 286, 321–332.

Yu, Y., Zhao, F., Yue, Y., Zhao, Y., & Zhou, D.X. (2024). Lysine acetylation of histone acetyltransferase adaptor protein ADA2 is a mechanism of metabolic control of chromatin modification in plants. Nature Plants, 10, 439–452.

Zhang, D., Zhang, D., Zhang, Y., Li, G., Sun, D., Zhou, B., & Li, J. (2024). Insights into the epigenetic basis of plant salt tolerance. *International Journal of Molecular Sciences*, *25*(21), 11698.

Zhang, N., Liu, B., Ma, C., Zhang, G., Chang, J., Si, H., & Wang, D. (2014). Transcriptome characterization and sequencing-based identification of drought-responsive genes in potato. *Molecular Biology Reports, 41*, 505–517.

Zhang, P., He, Y., & Huang, S. (2024). Unlocking epigenetic breeding potential in tomato and potato. *Abiotech*, *5*(4), 507-518.

Zhu, J. K. (2001). Plant salt tolerance. *Trends in plant science*, *6*(2), 66-71.

Zhu, J. K. (2002). Salt and drought stress signal transduction in plants. *Annual review of plant biology*, *53*(1), 247-273.

CHAPTER II

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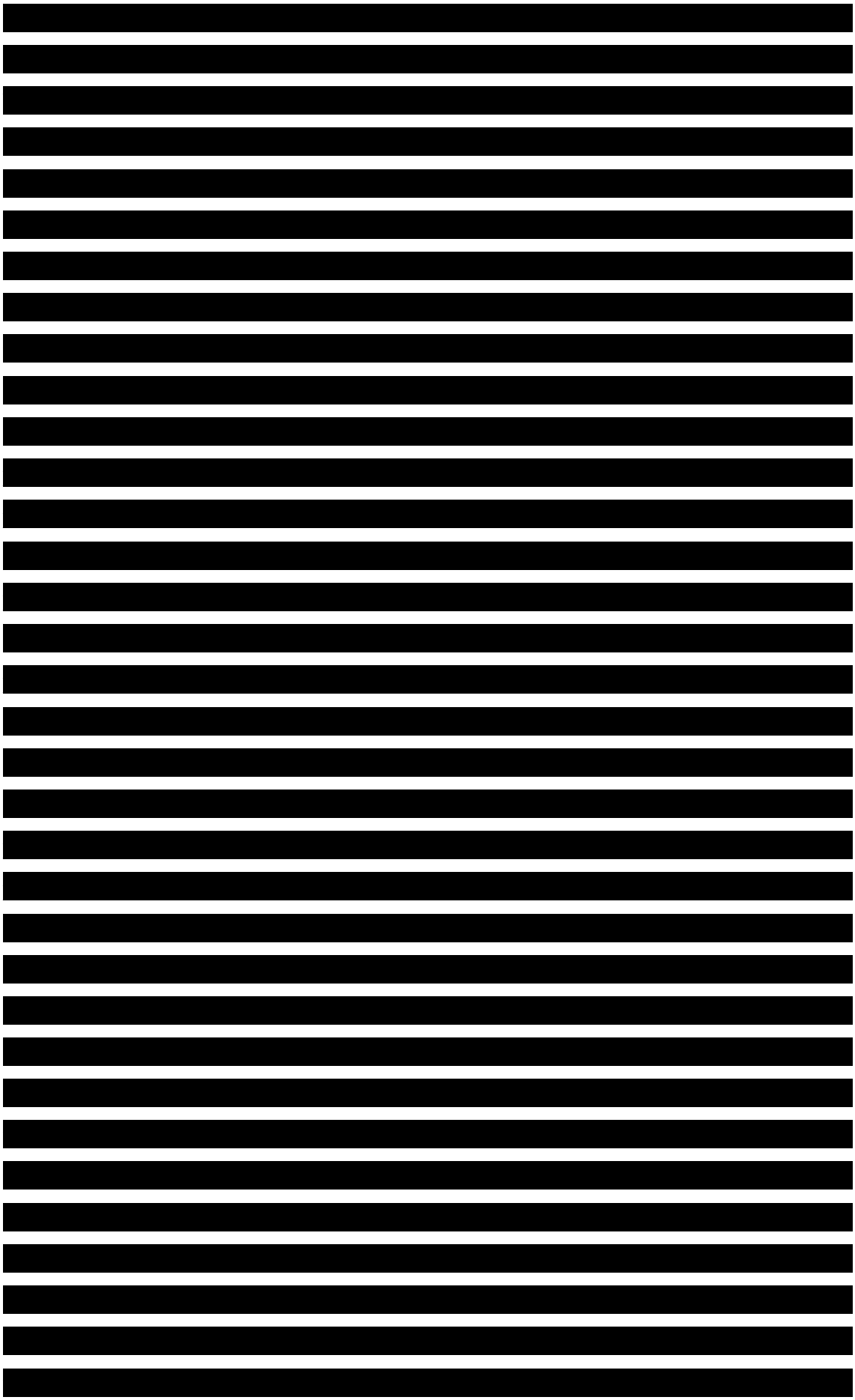

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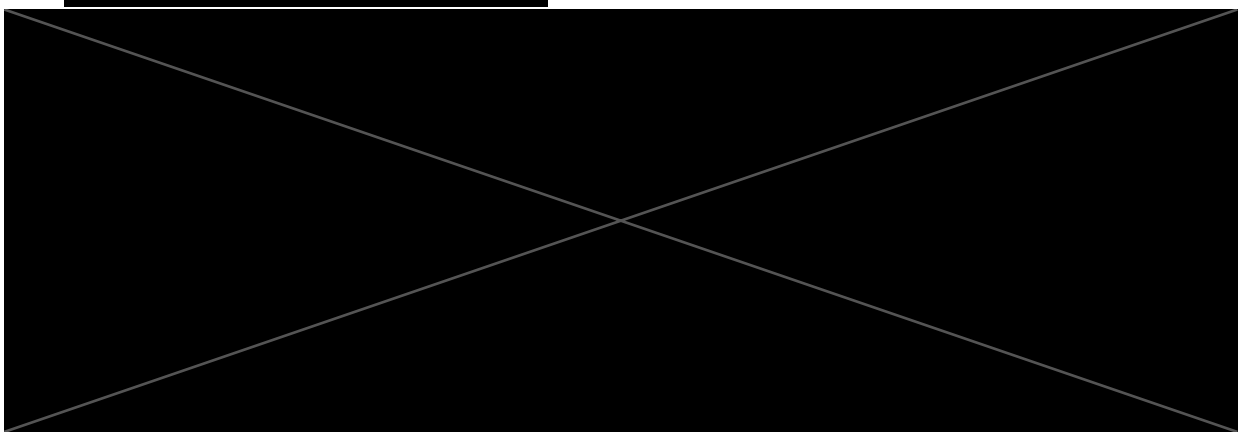

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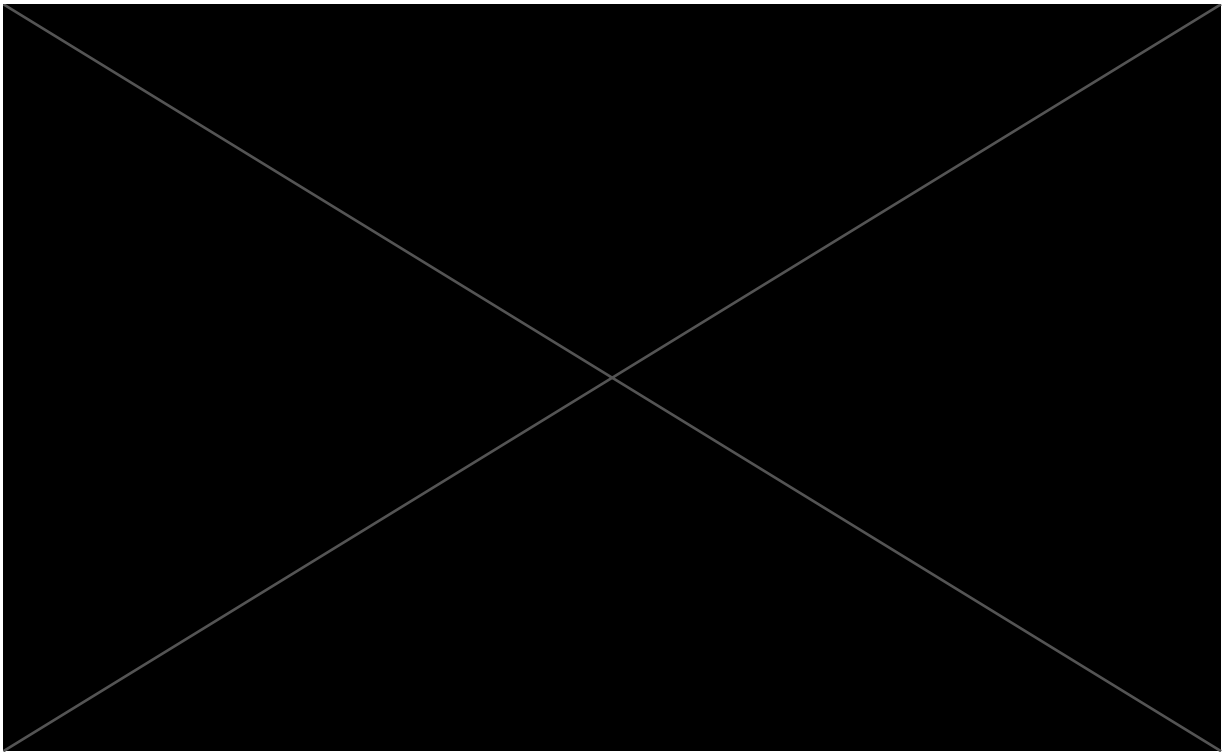

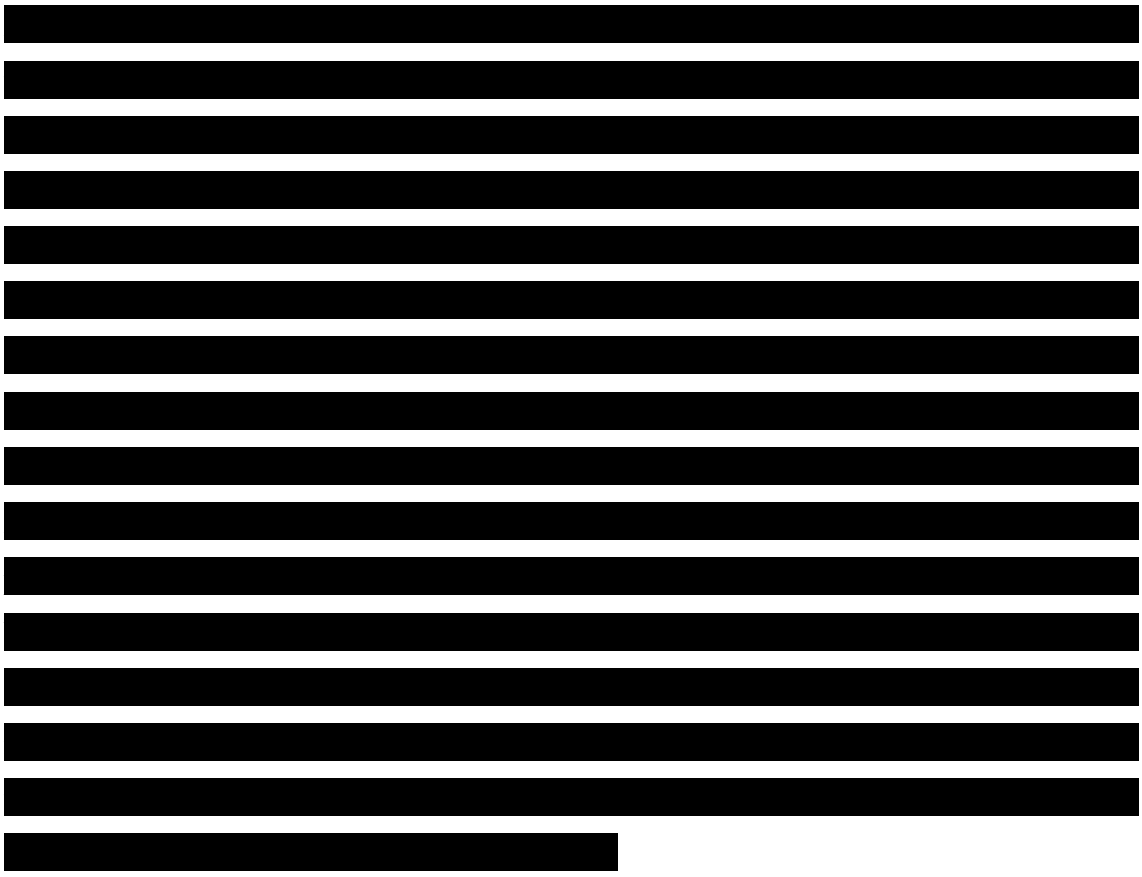

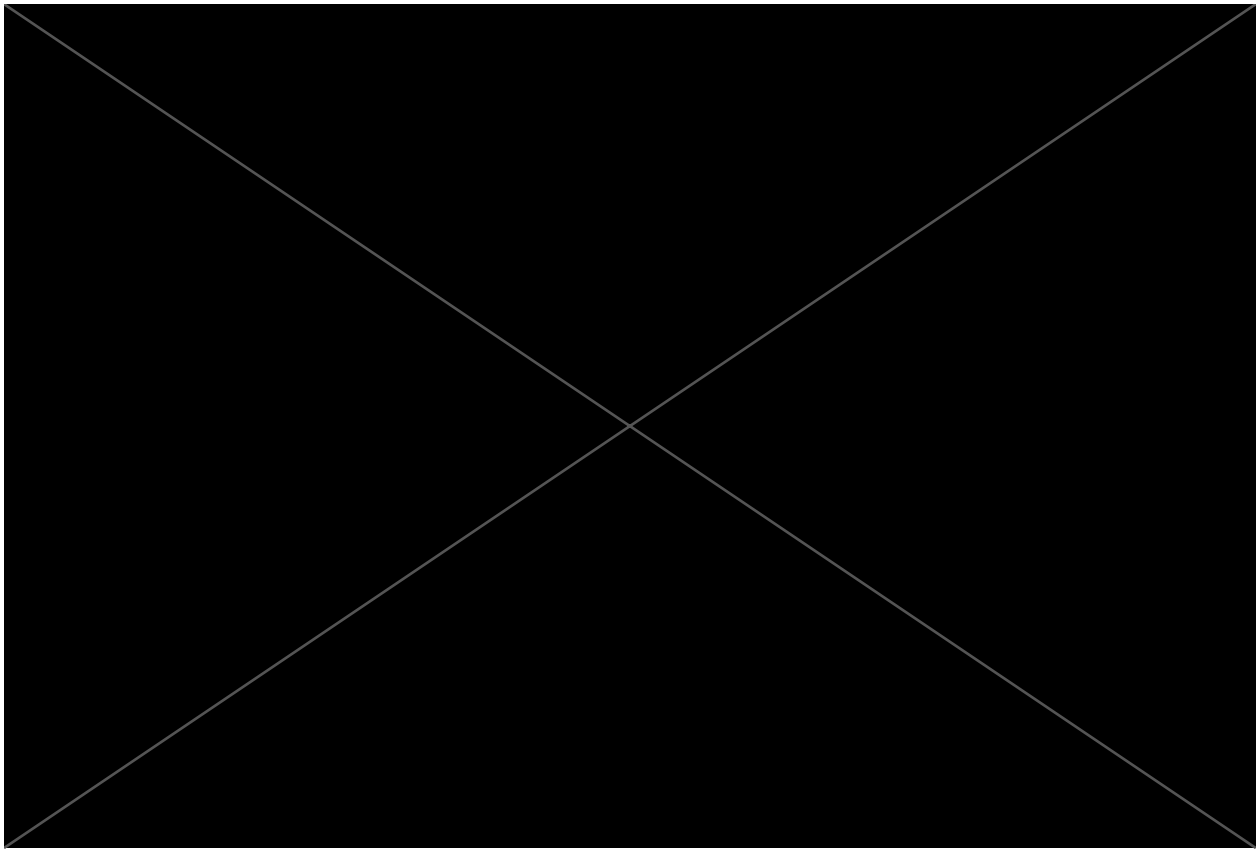

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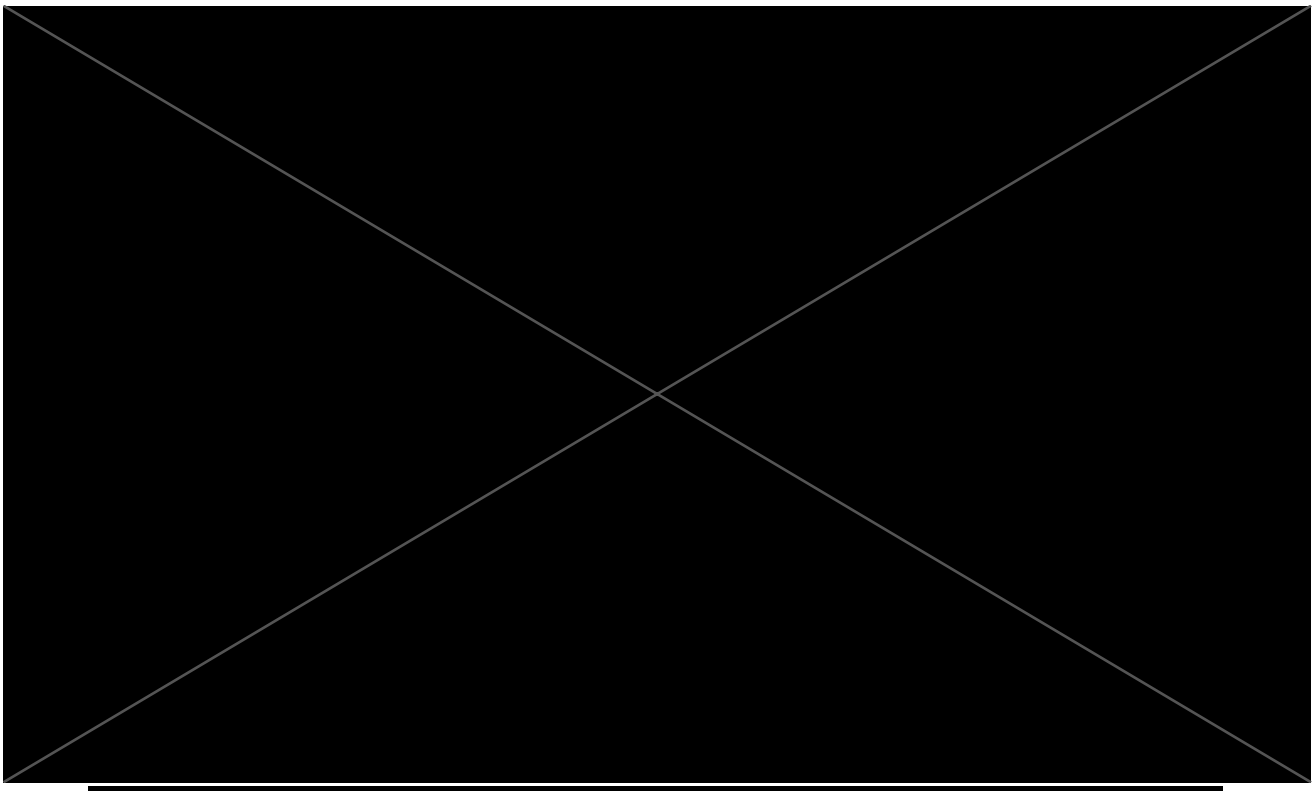

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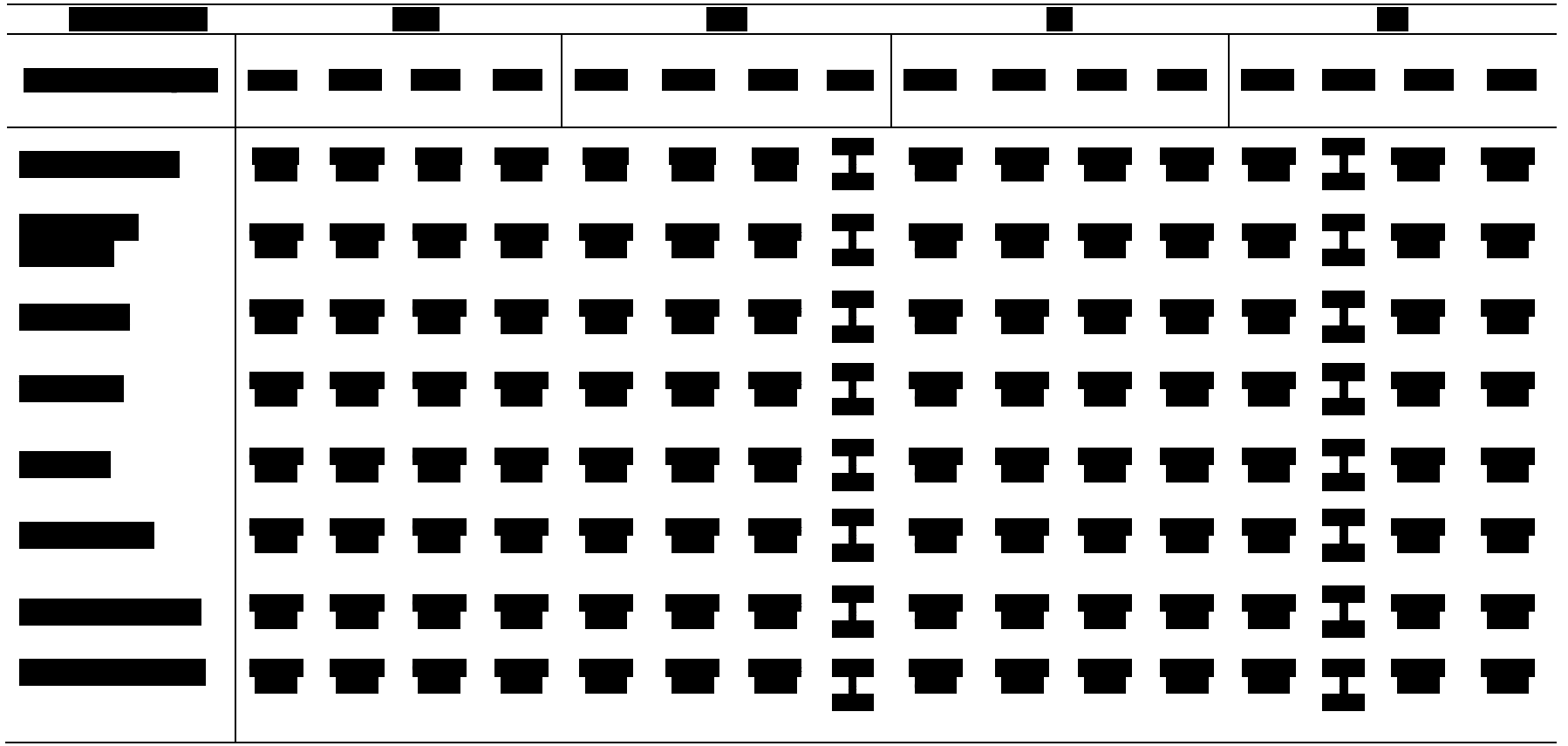

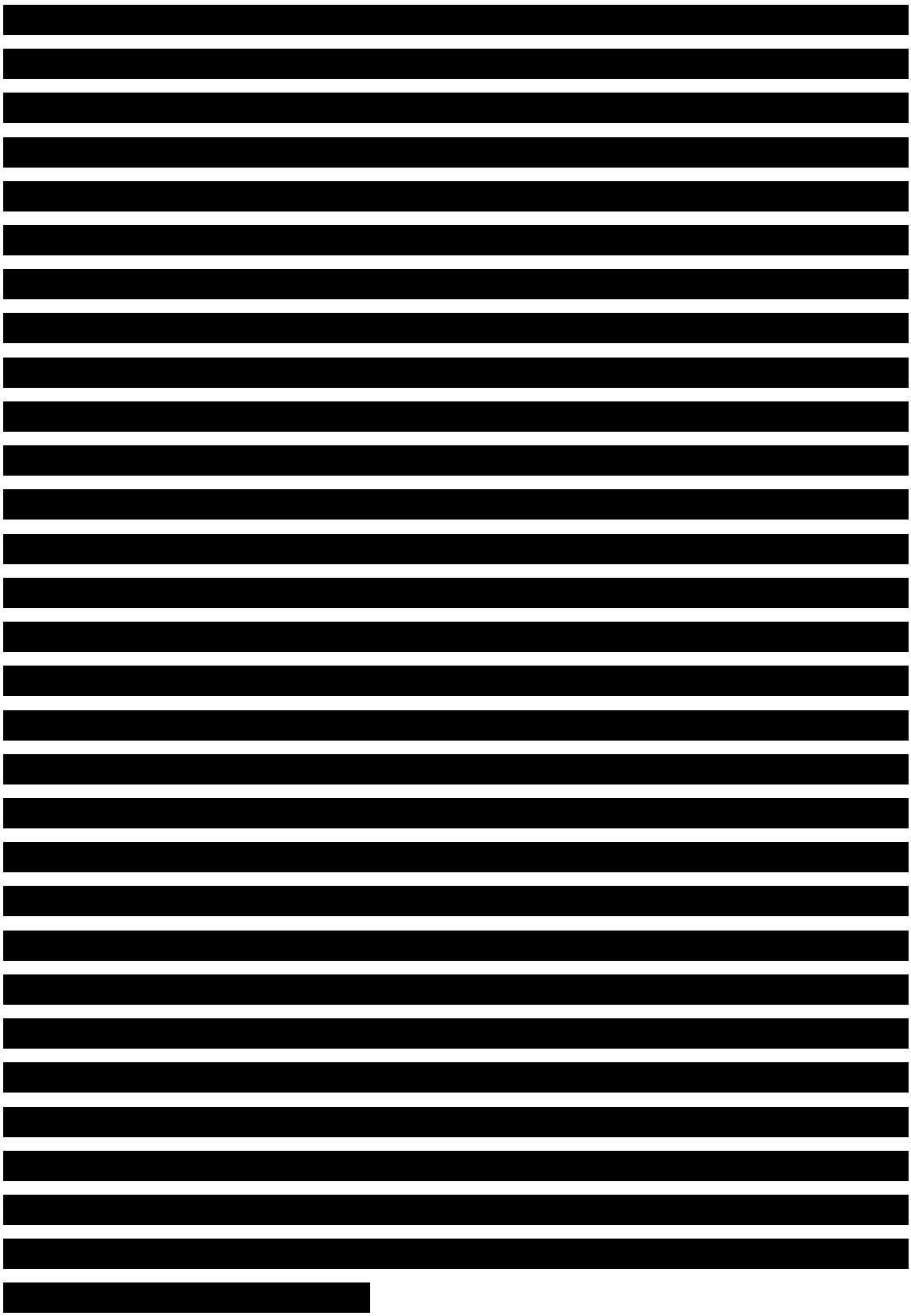

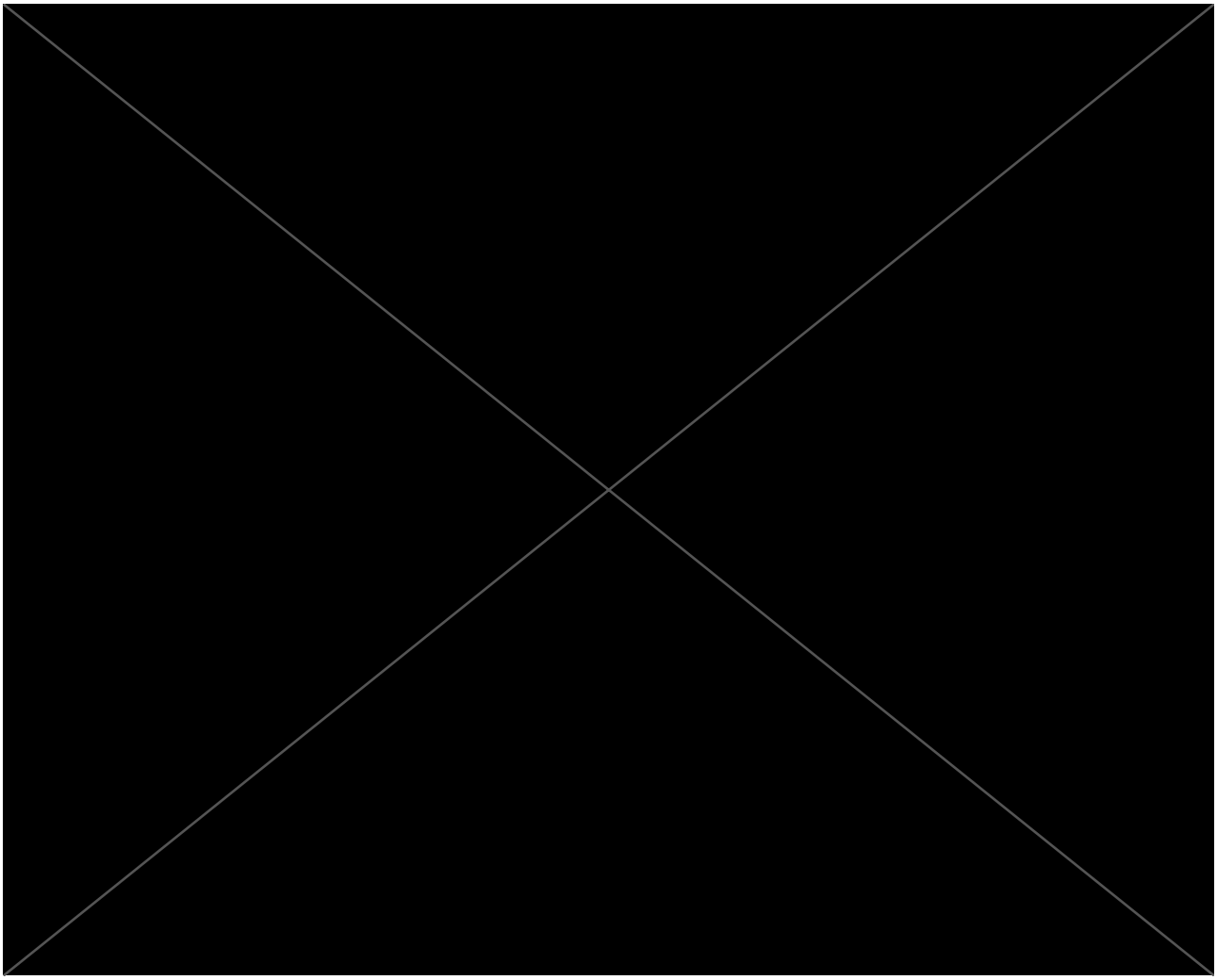

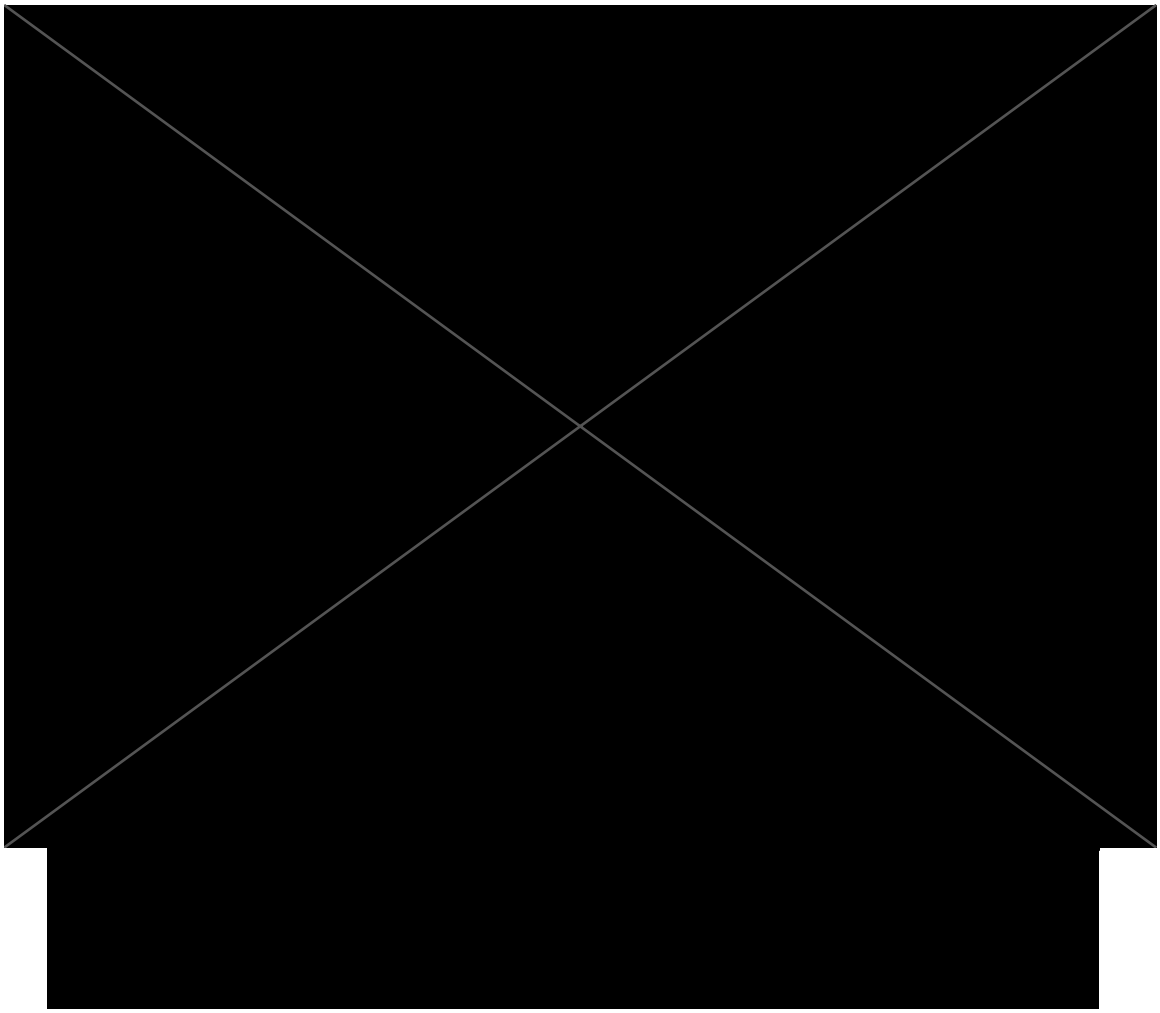

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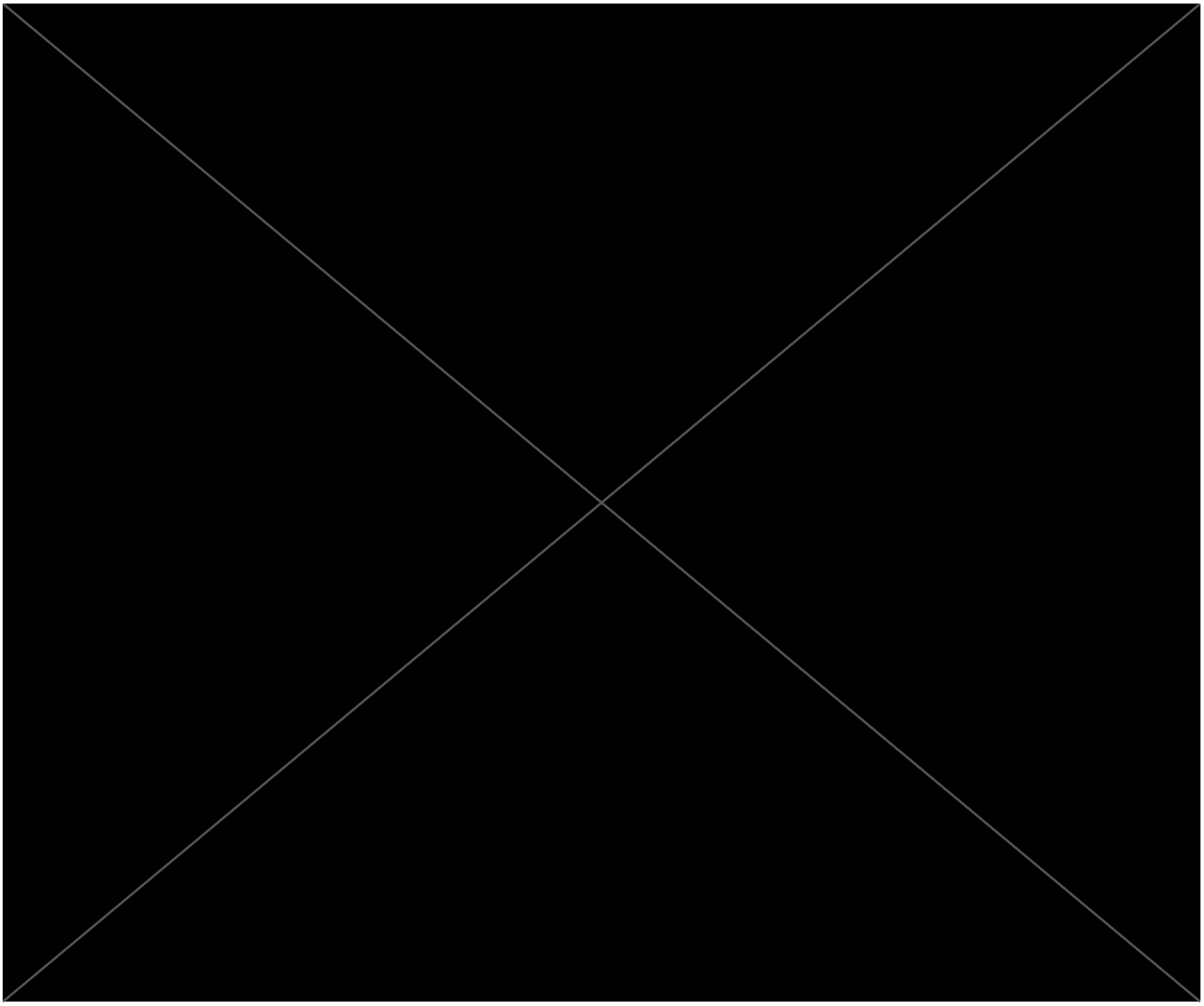

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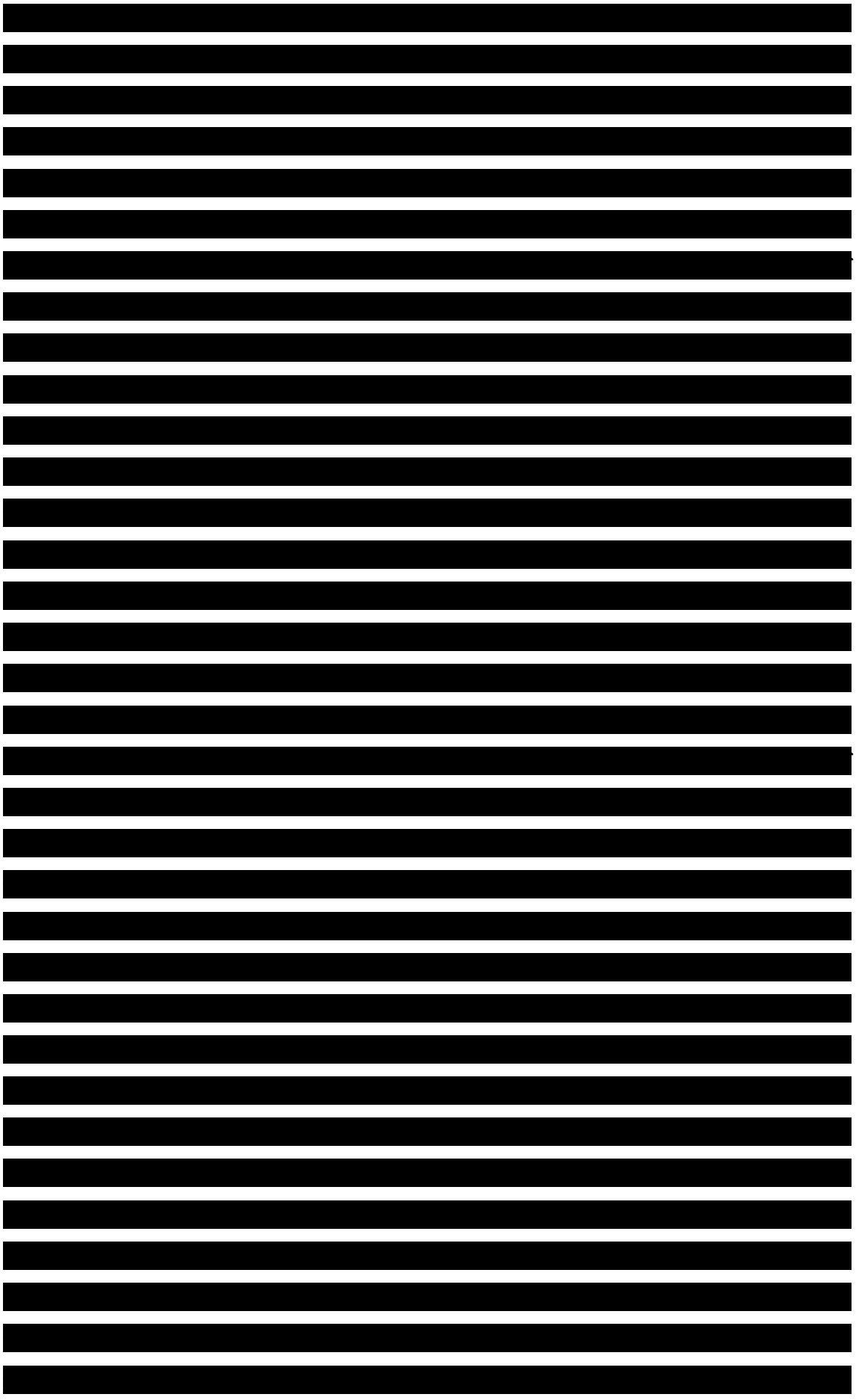

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D’Amelia, V., Piccinelli, A. L., Docimo, T., Cirillo, V., Maggio, A., Chiaiese, P., ... & Carputo, D. (2025). Expanded functional roles of R2R3-MYB (S6) transcription factors in balancing phenylpropanoid and phenolamide pathways in Solanaceae. Plant And Cell Physiology, pcaf028.

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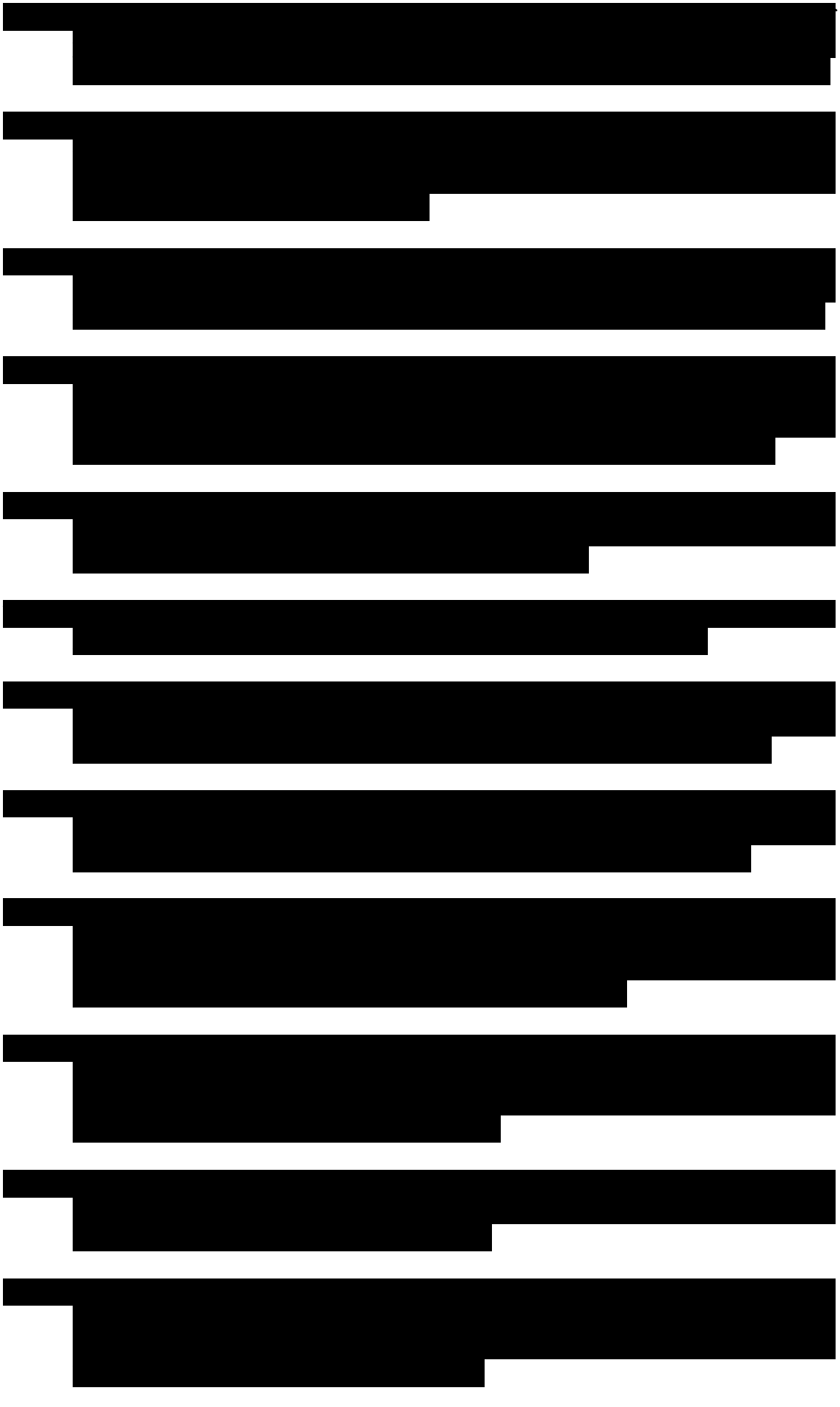

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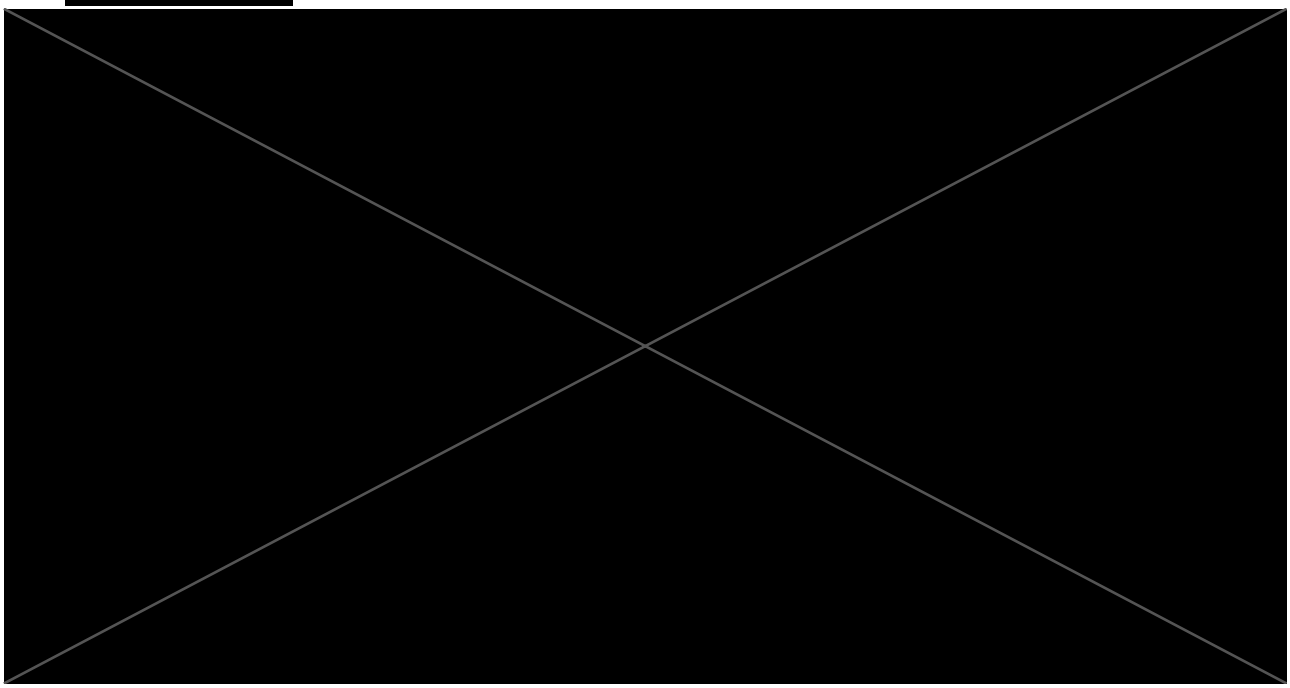

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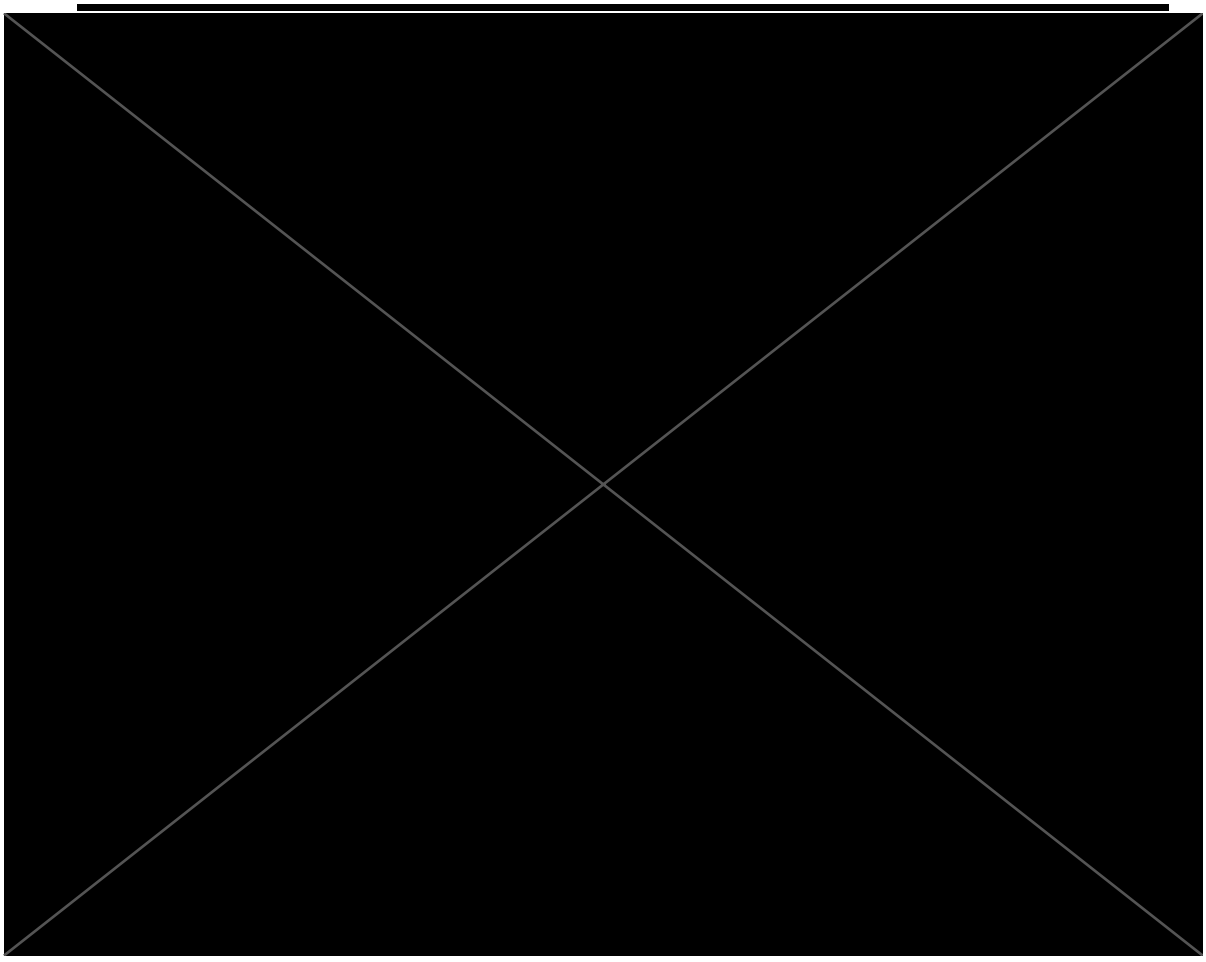

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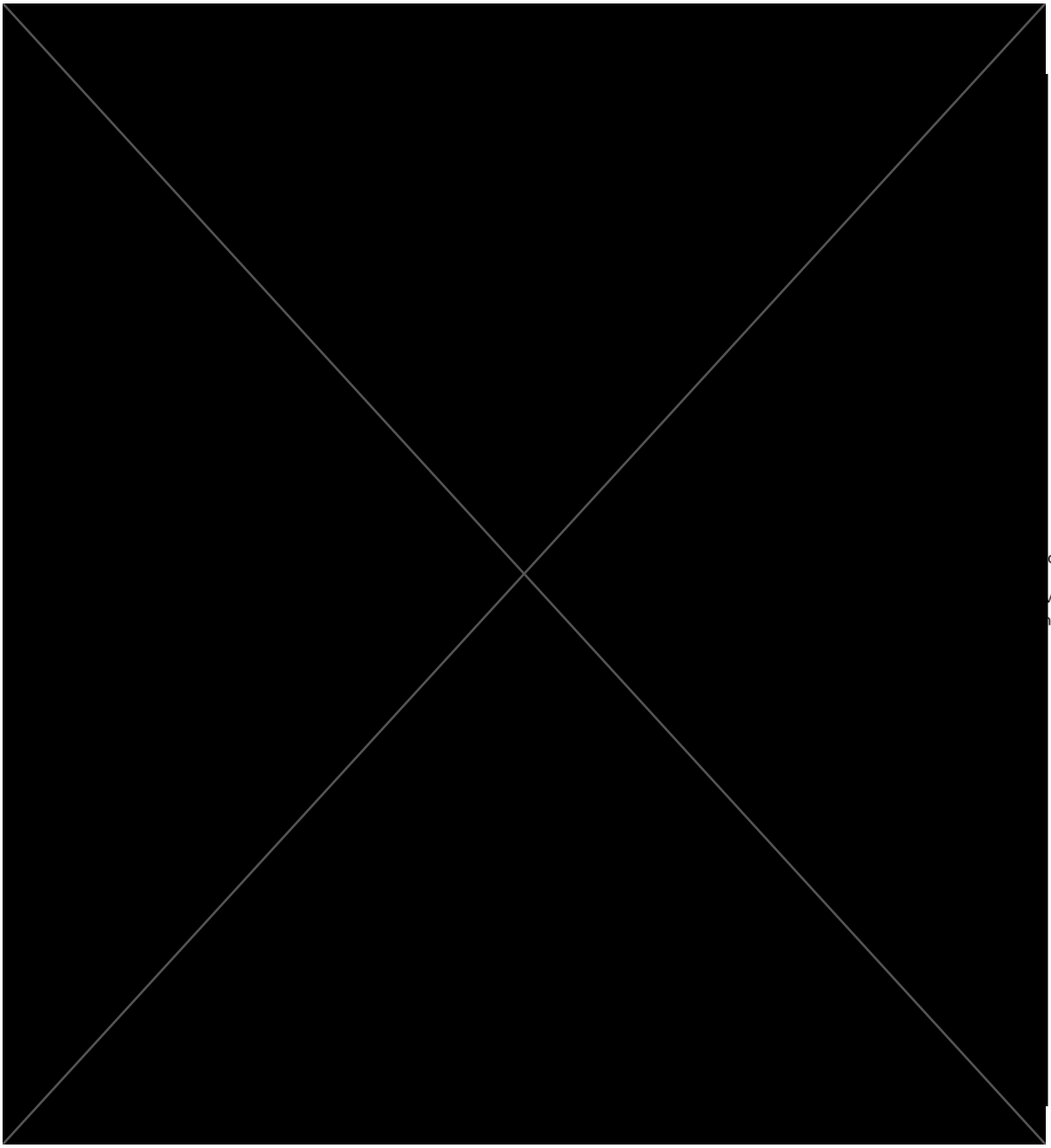

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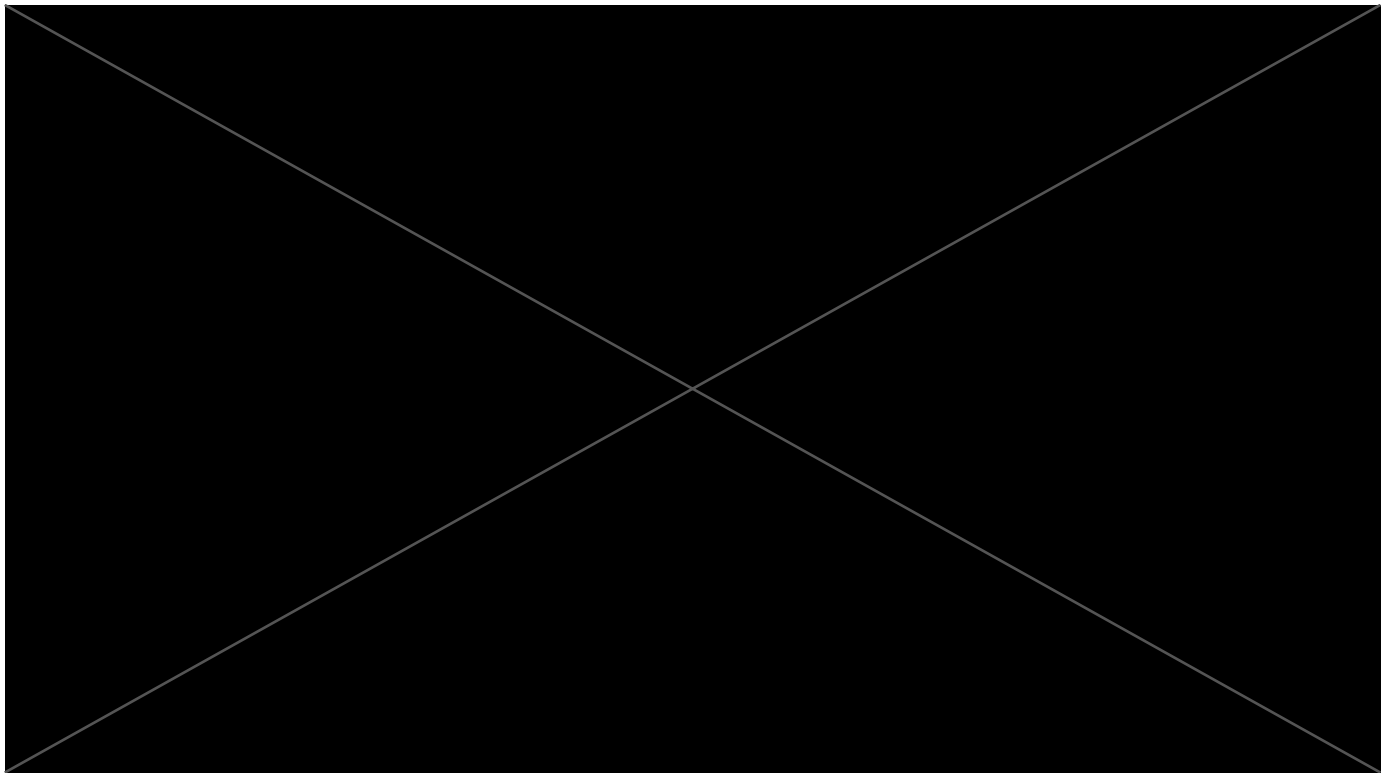

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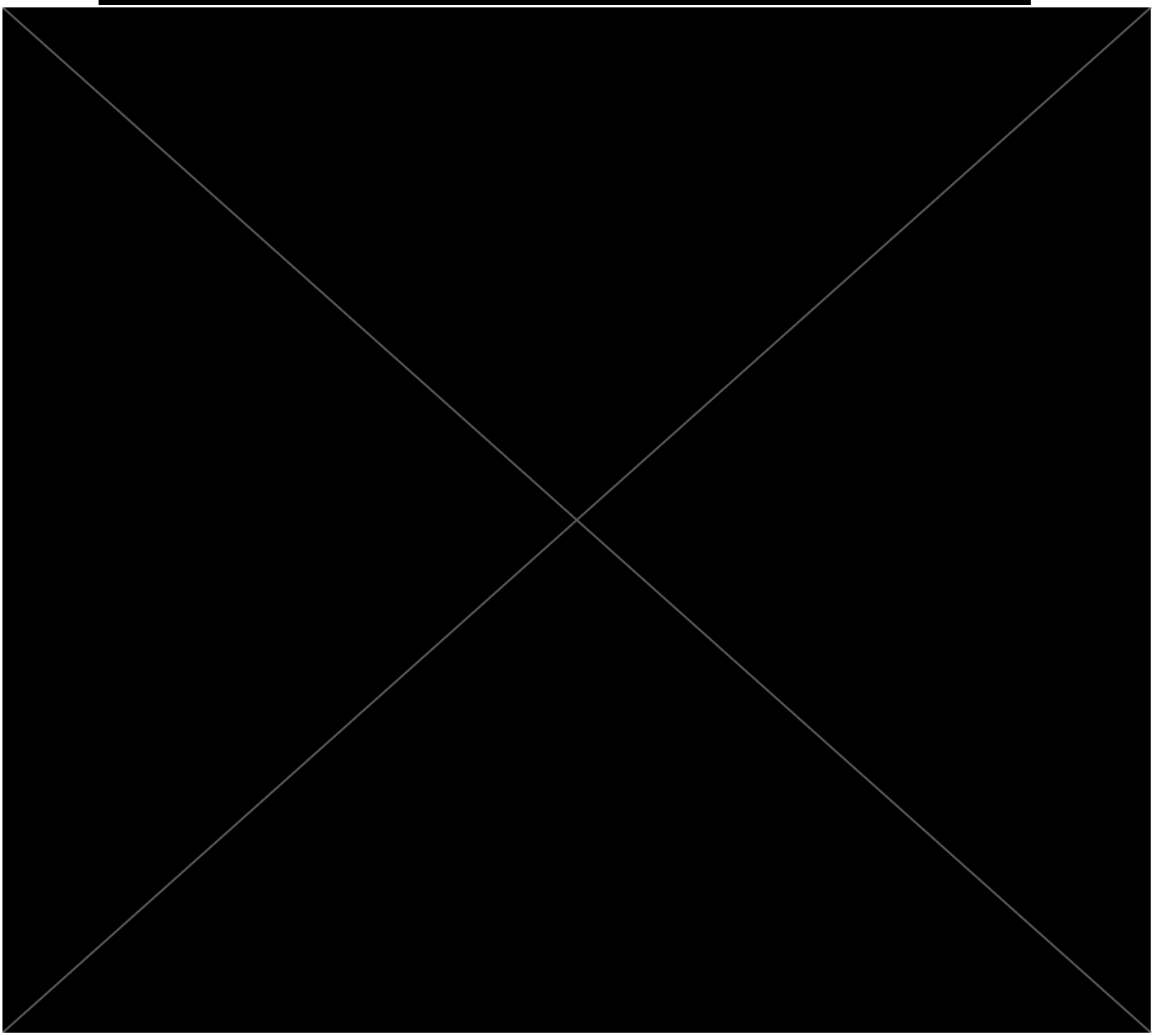

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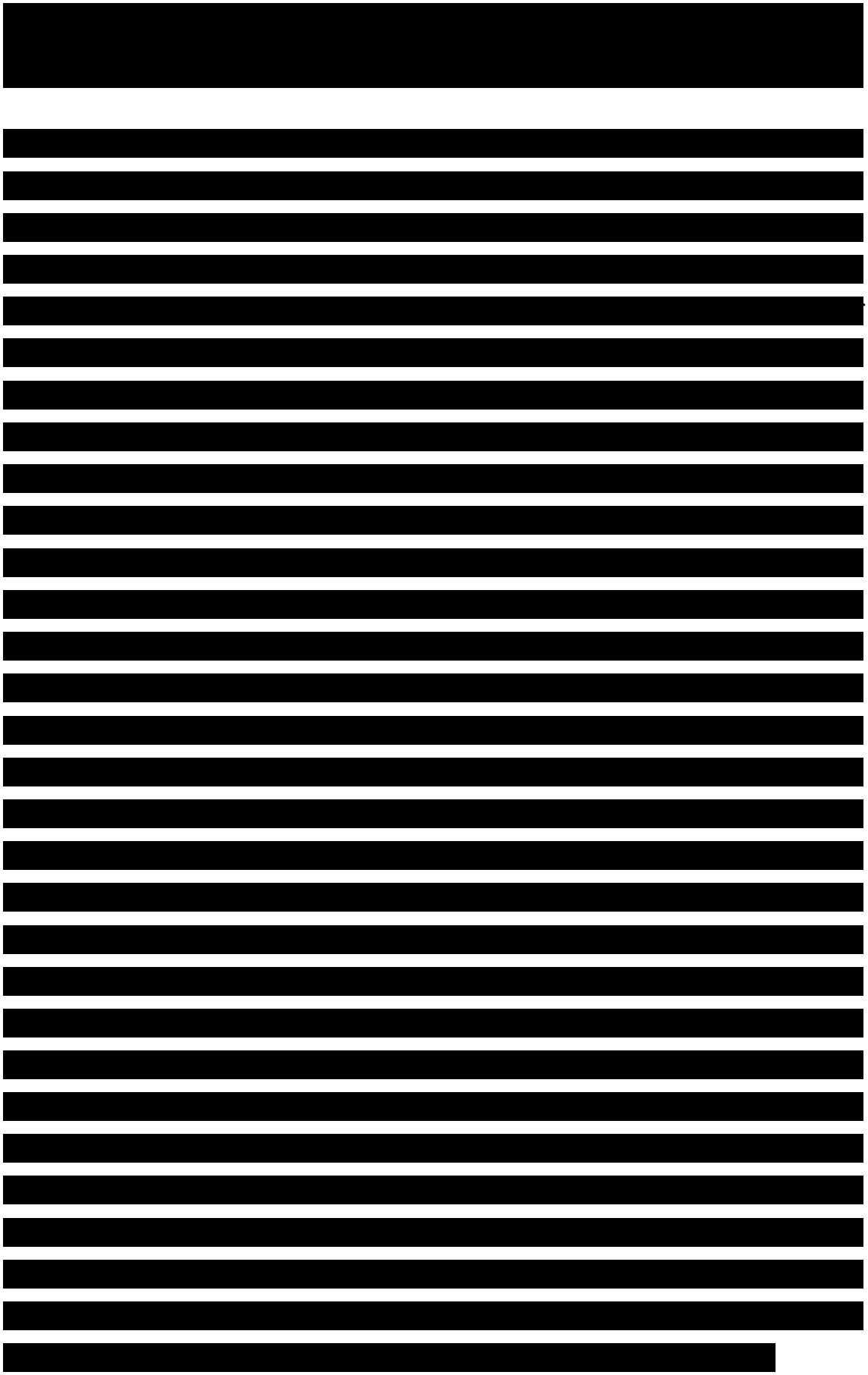

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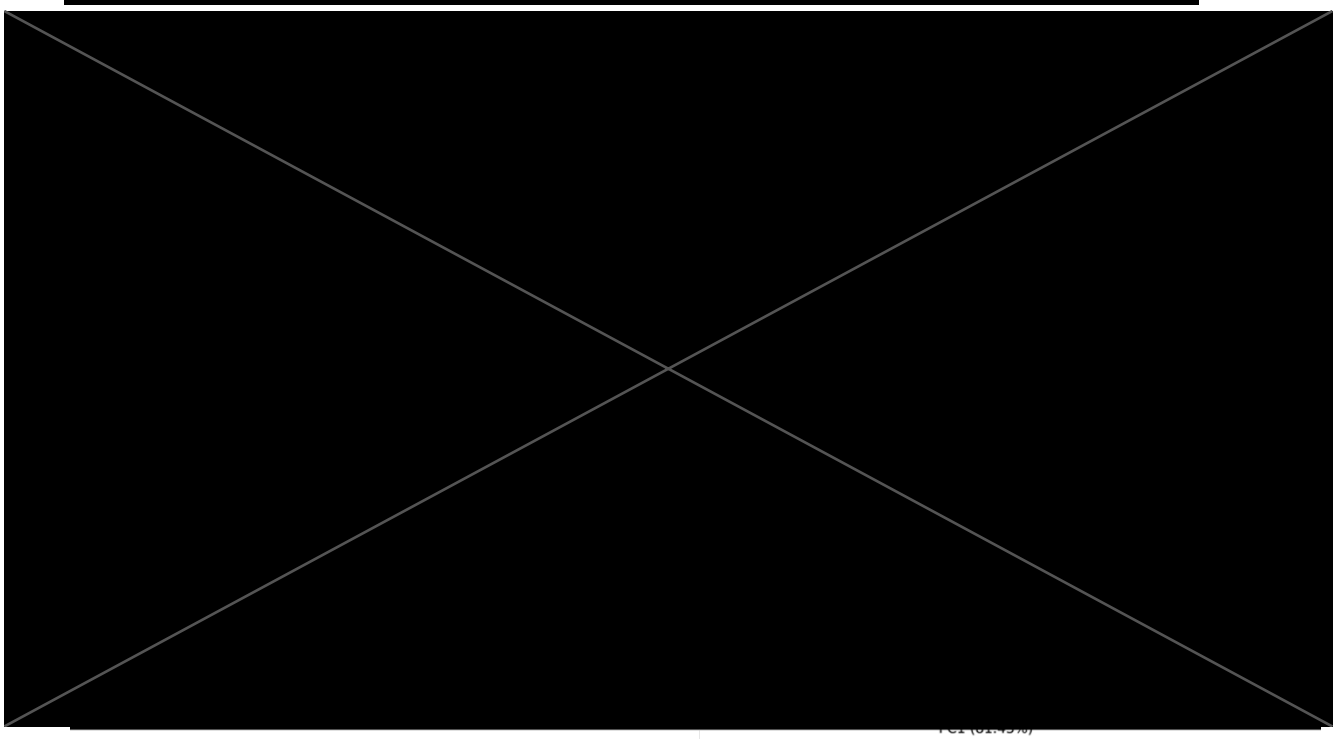

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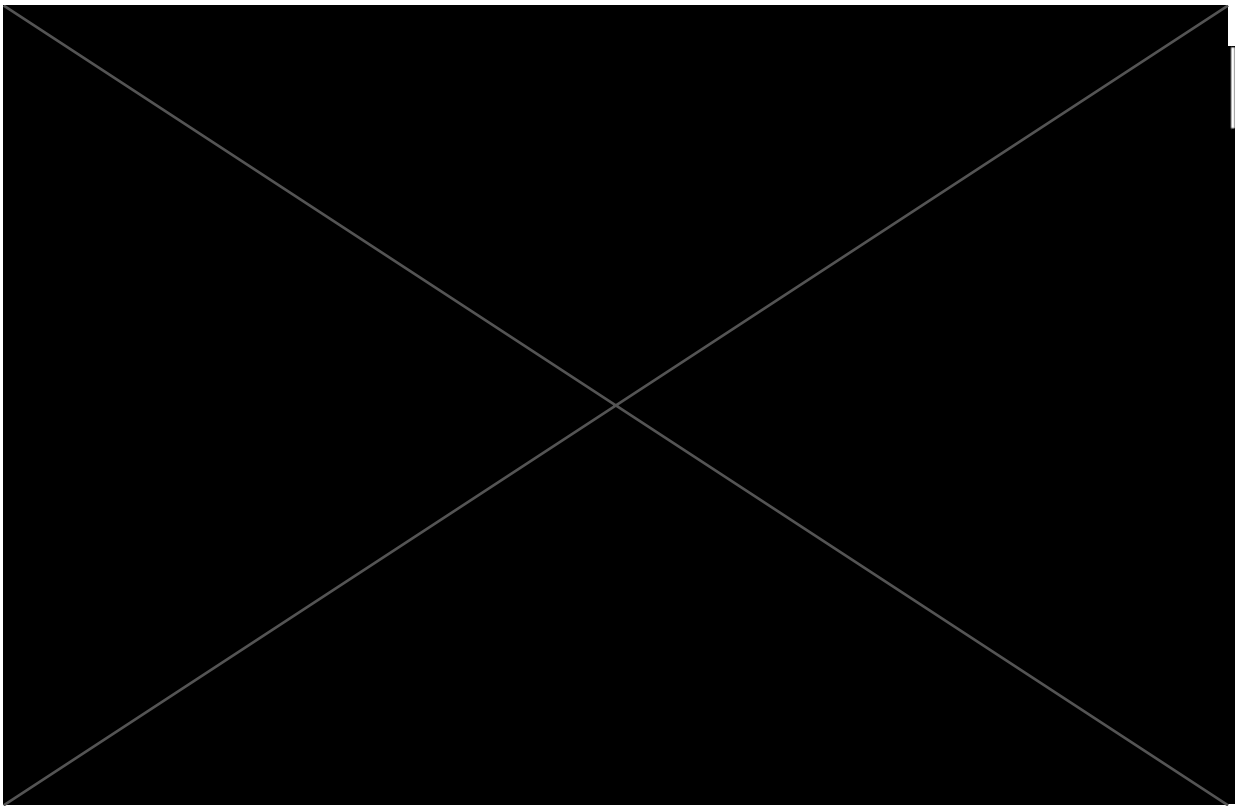

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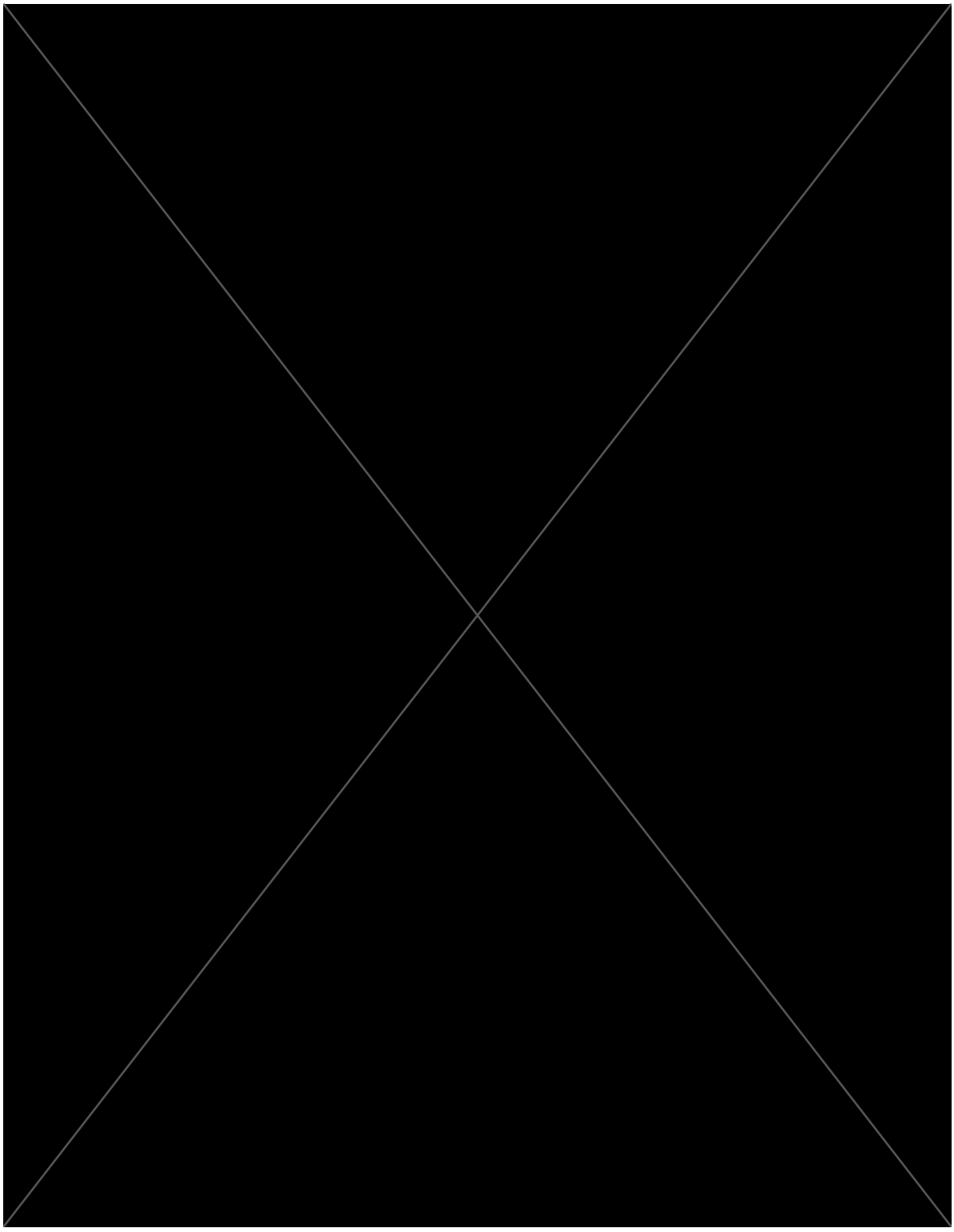

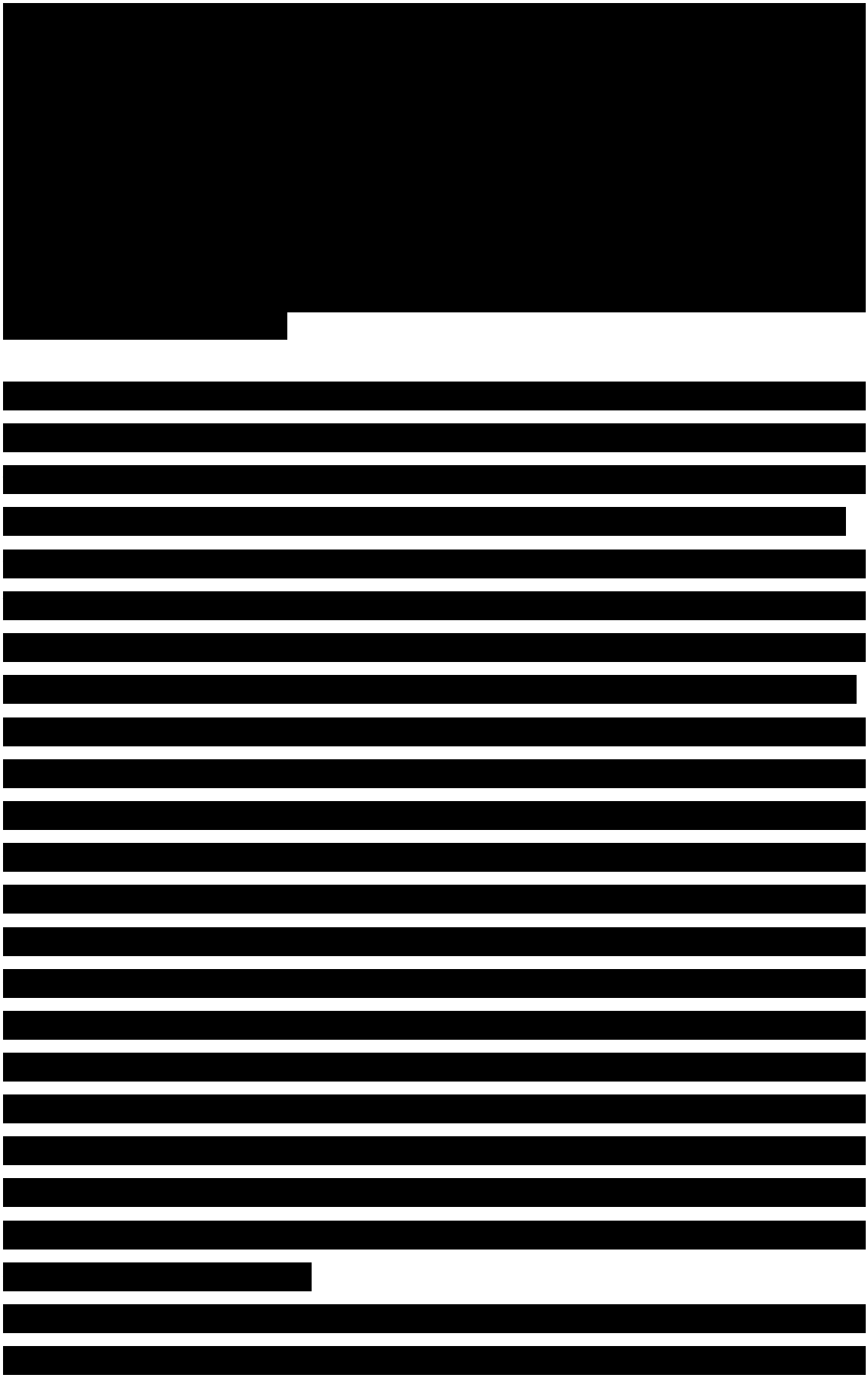

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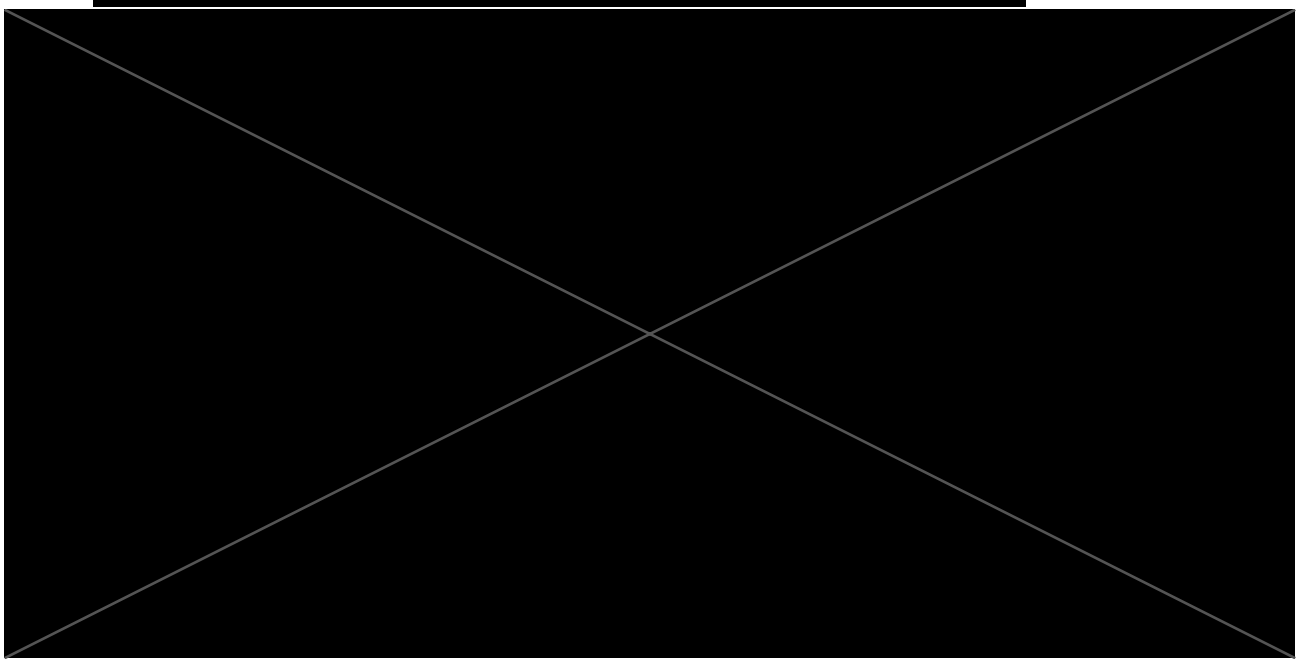

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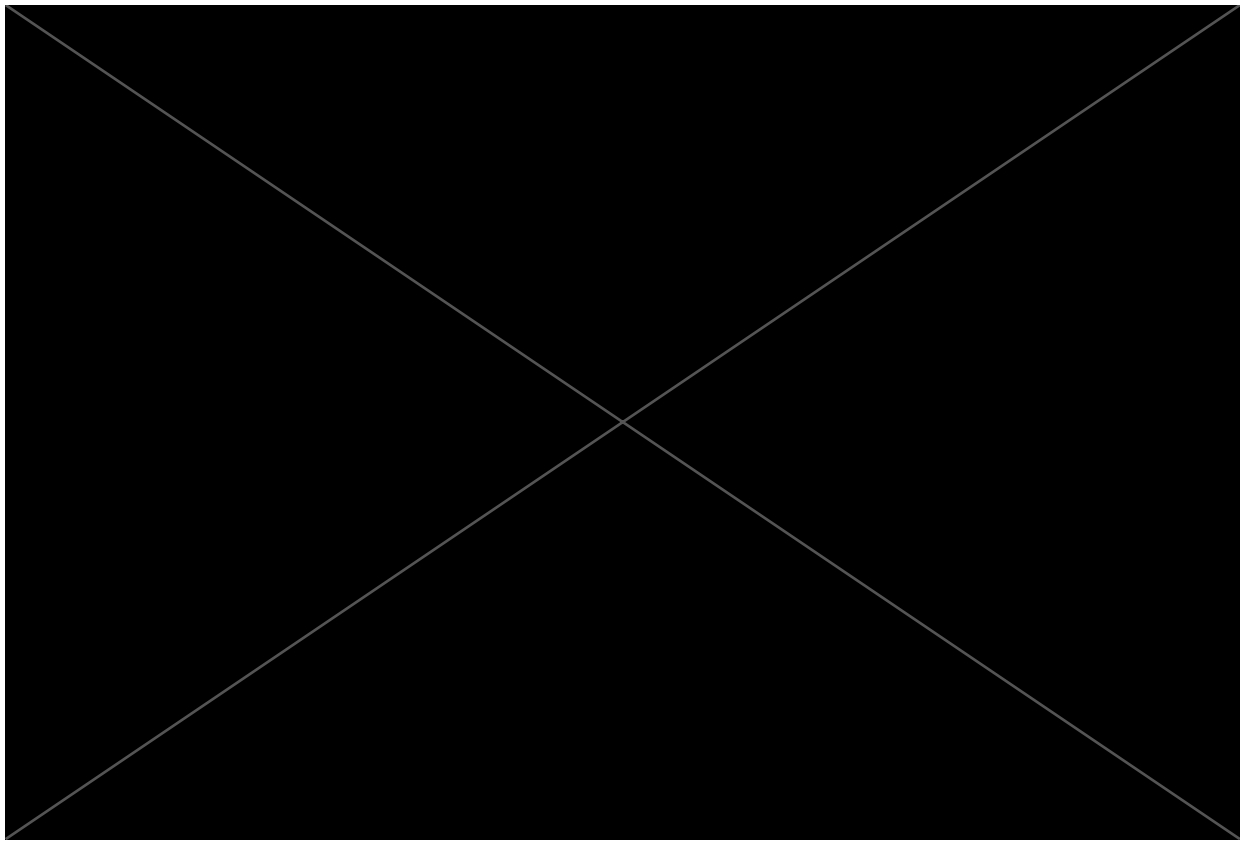

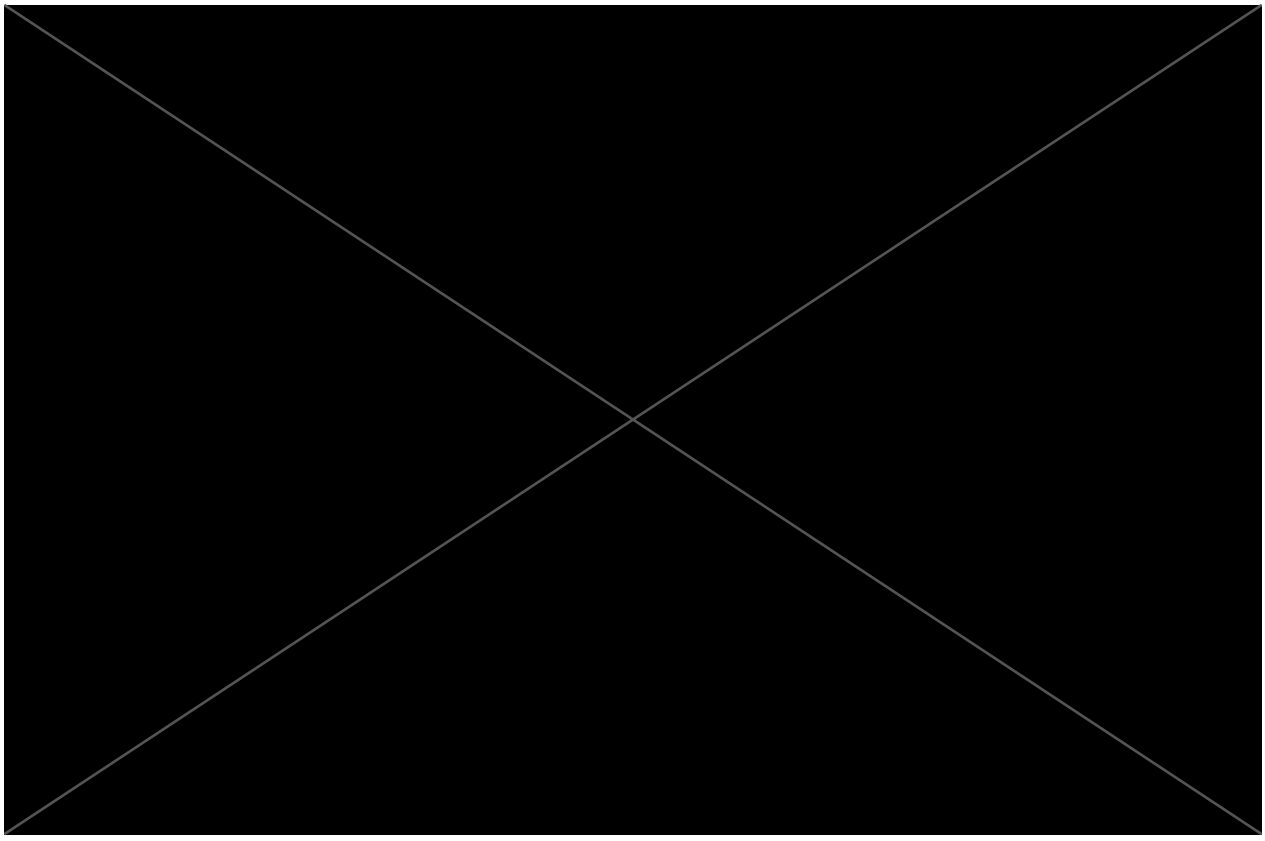

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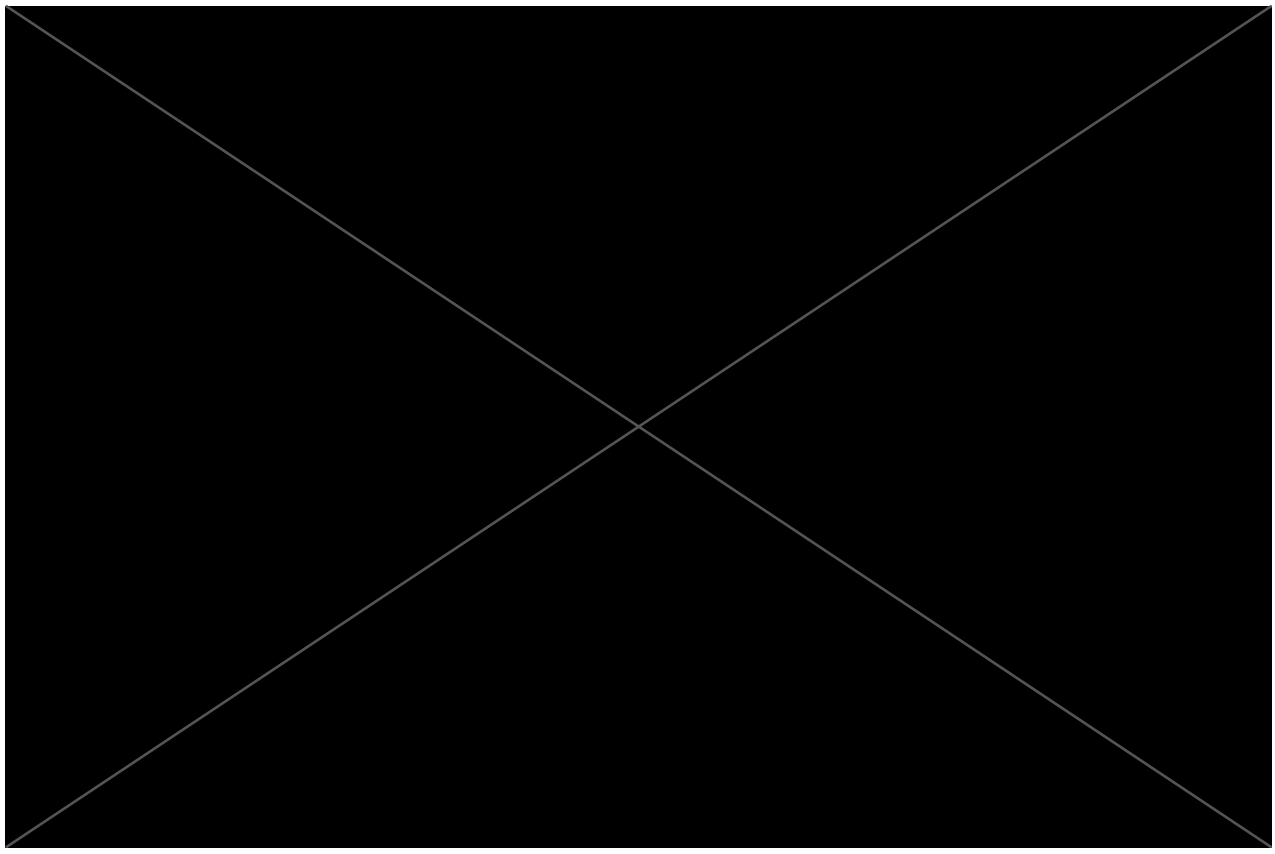

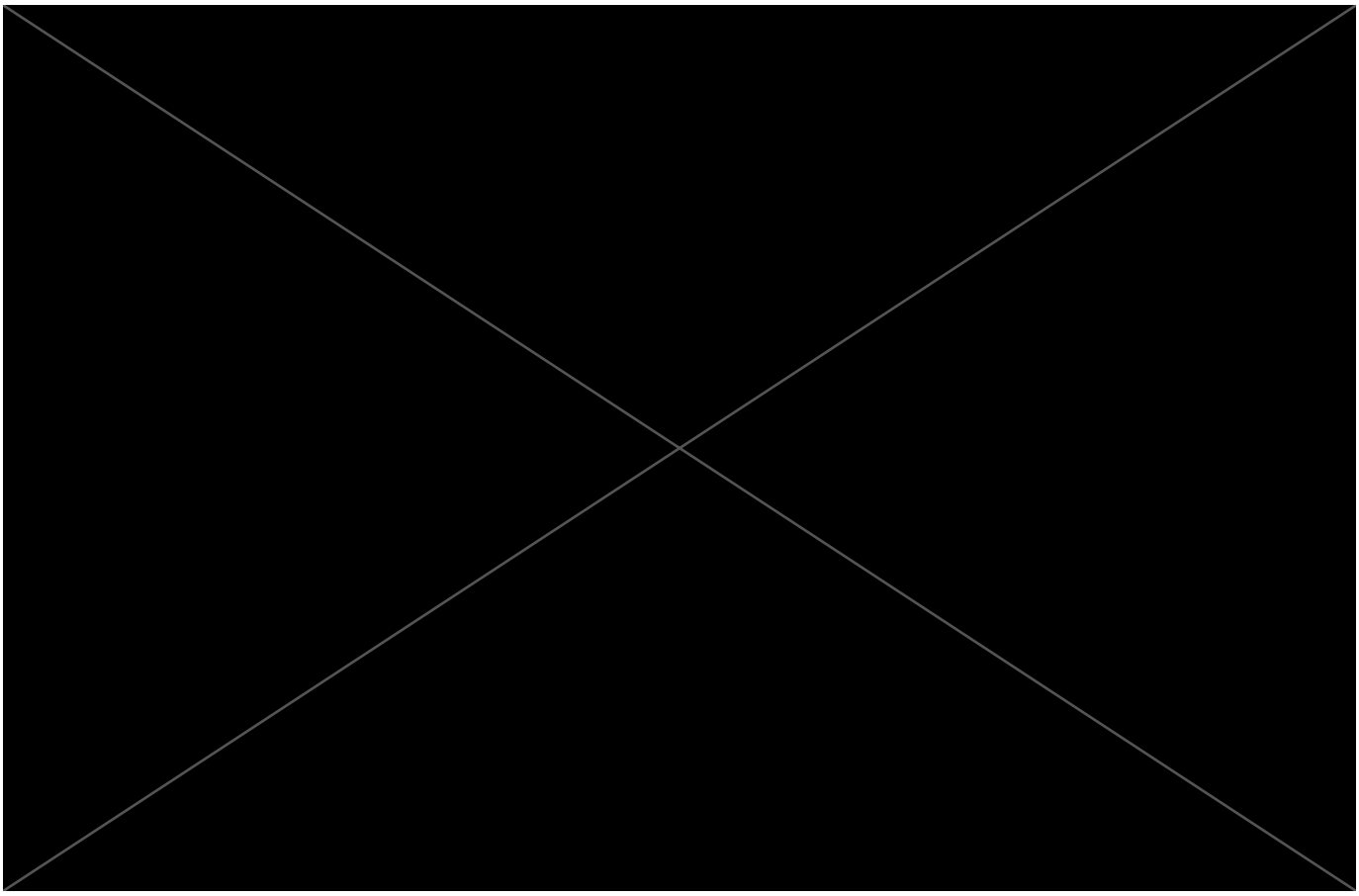

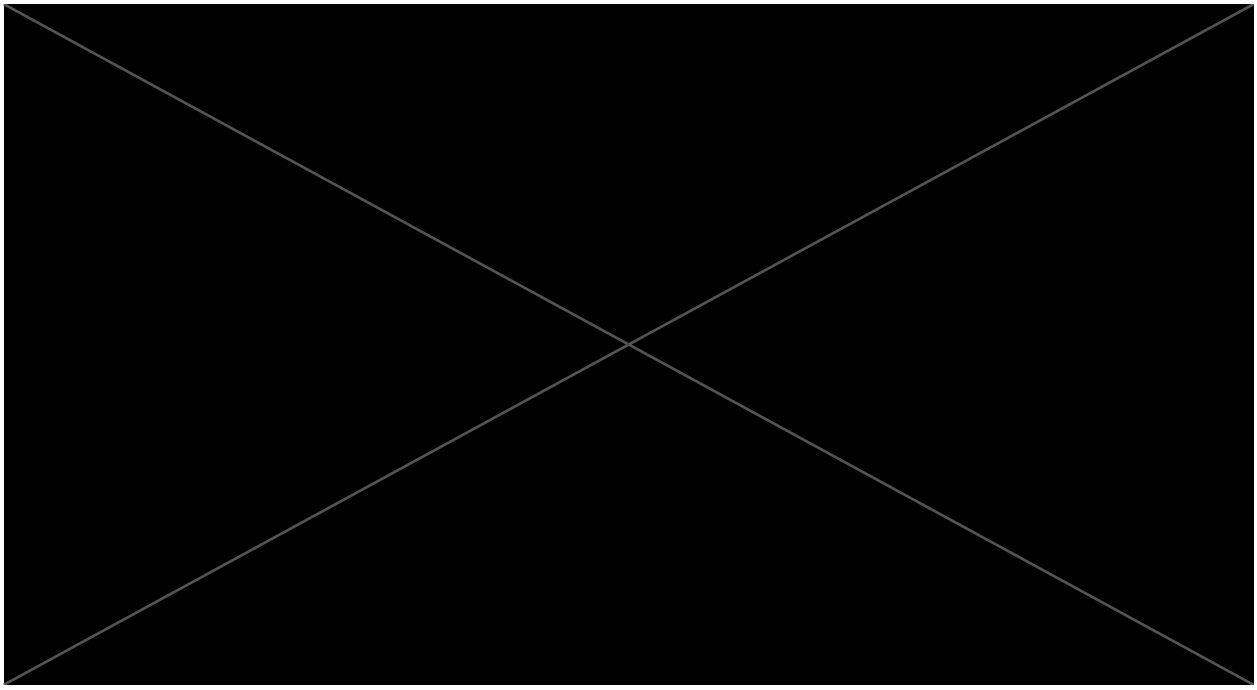

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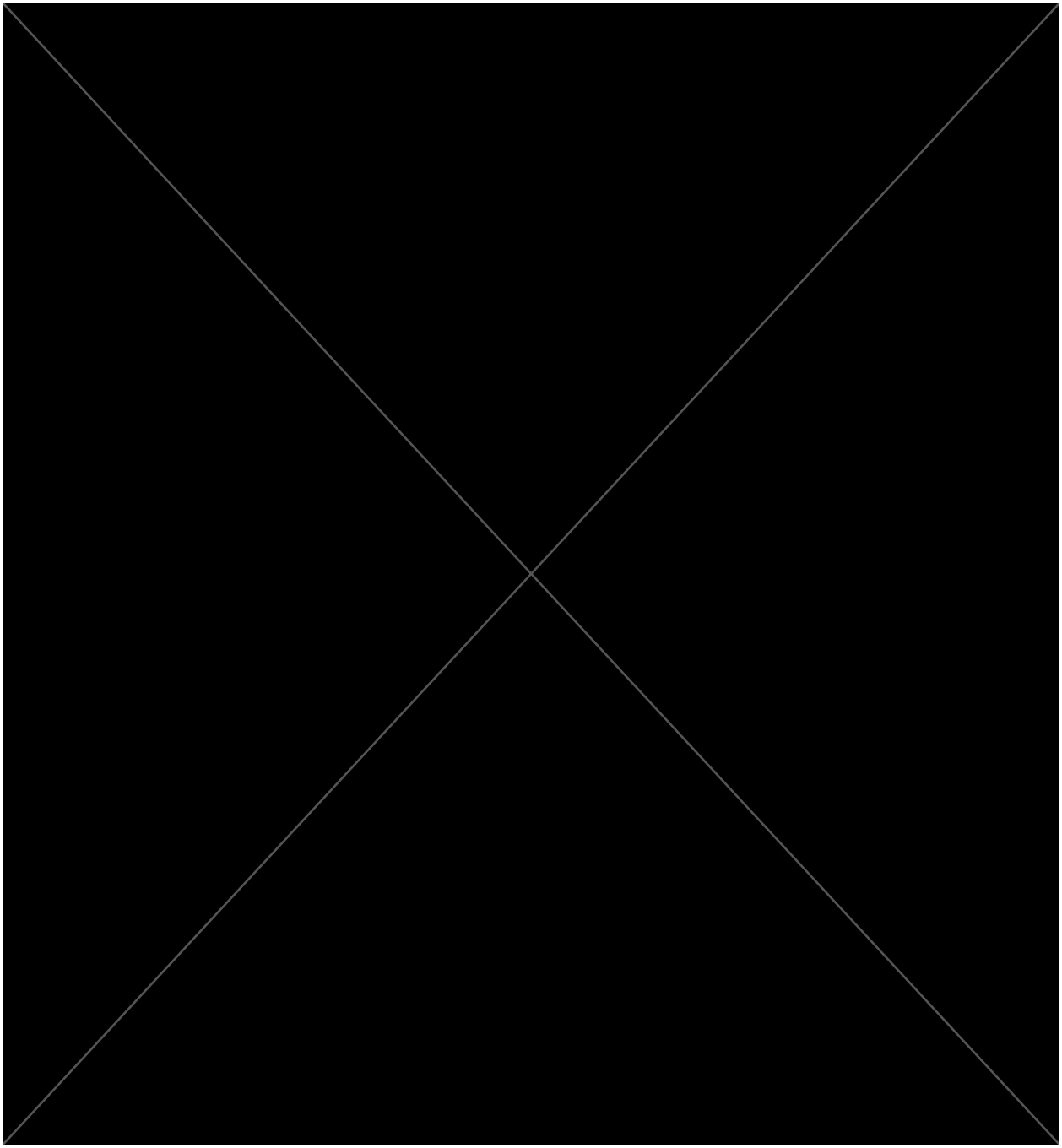

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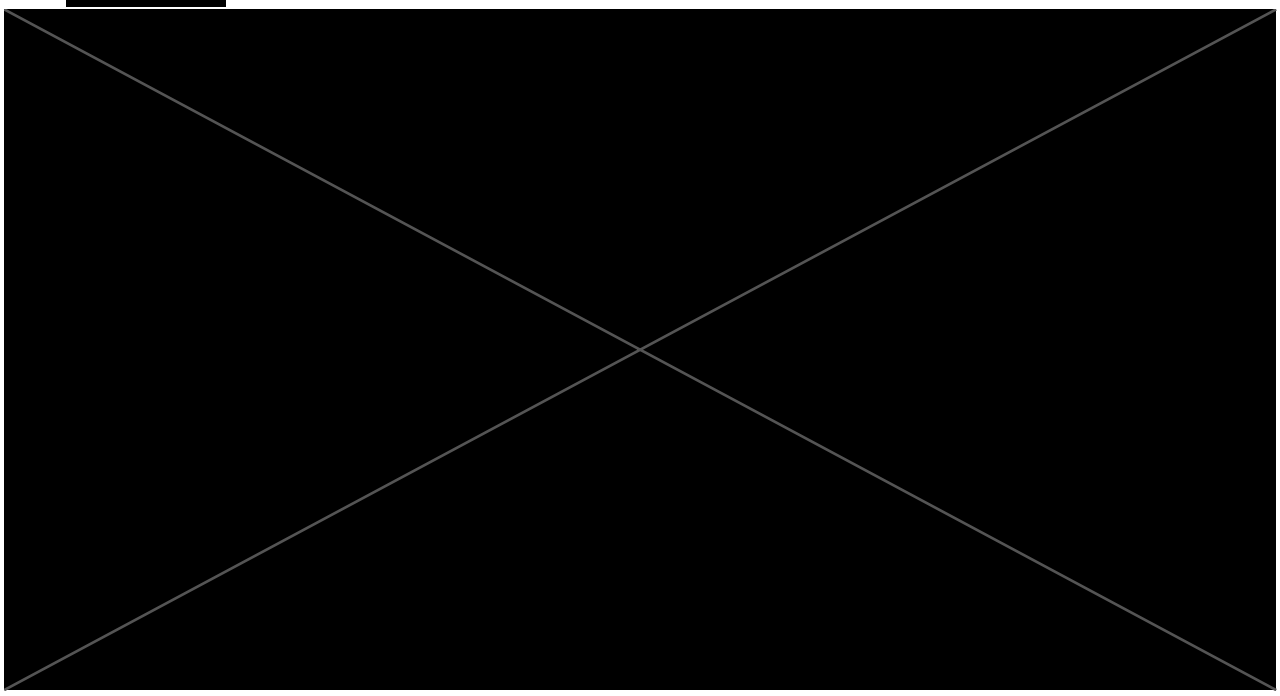

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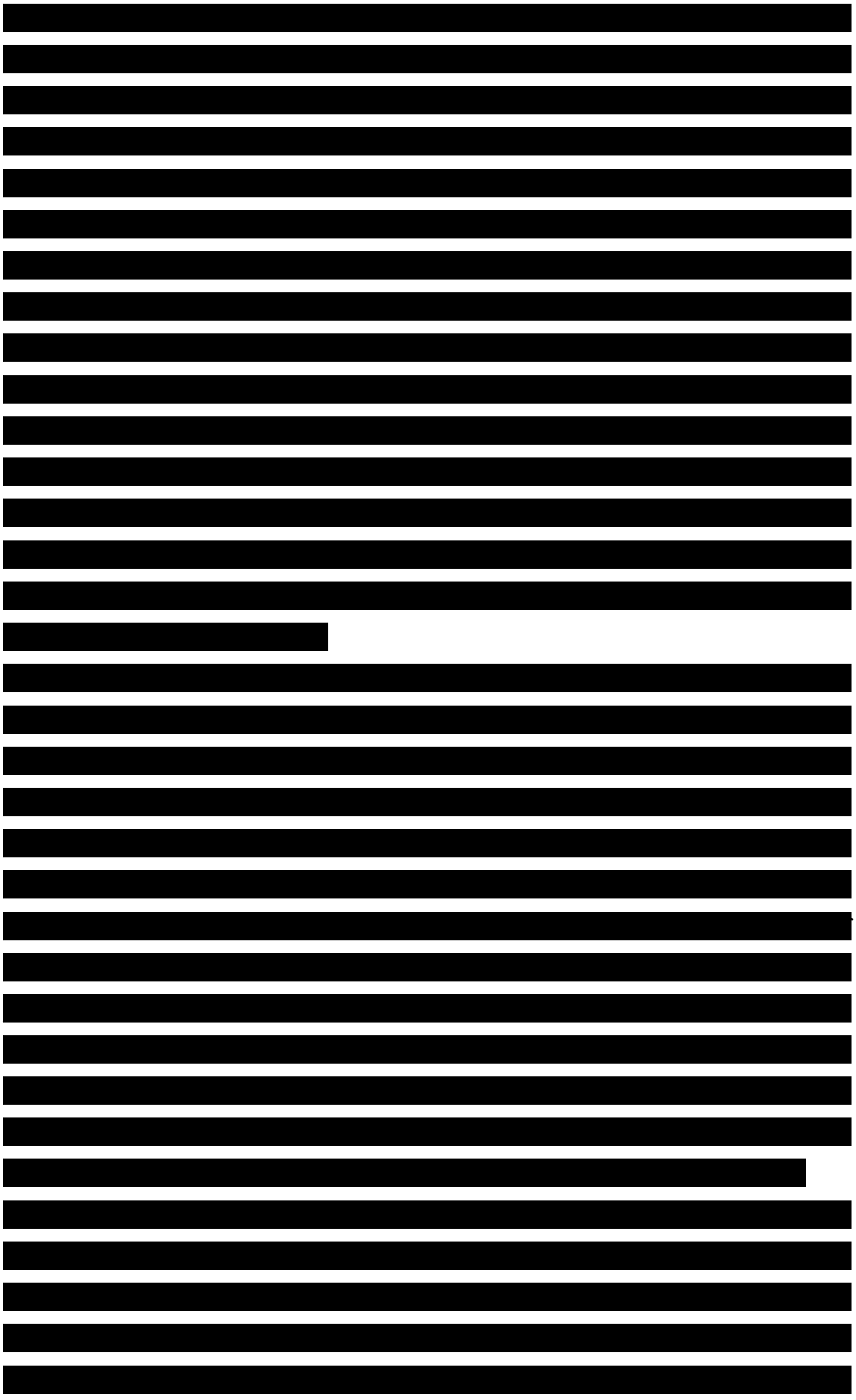

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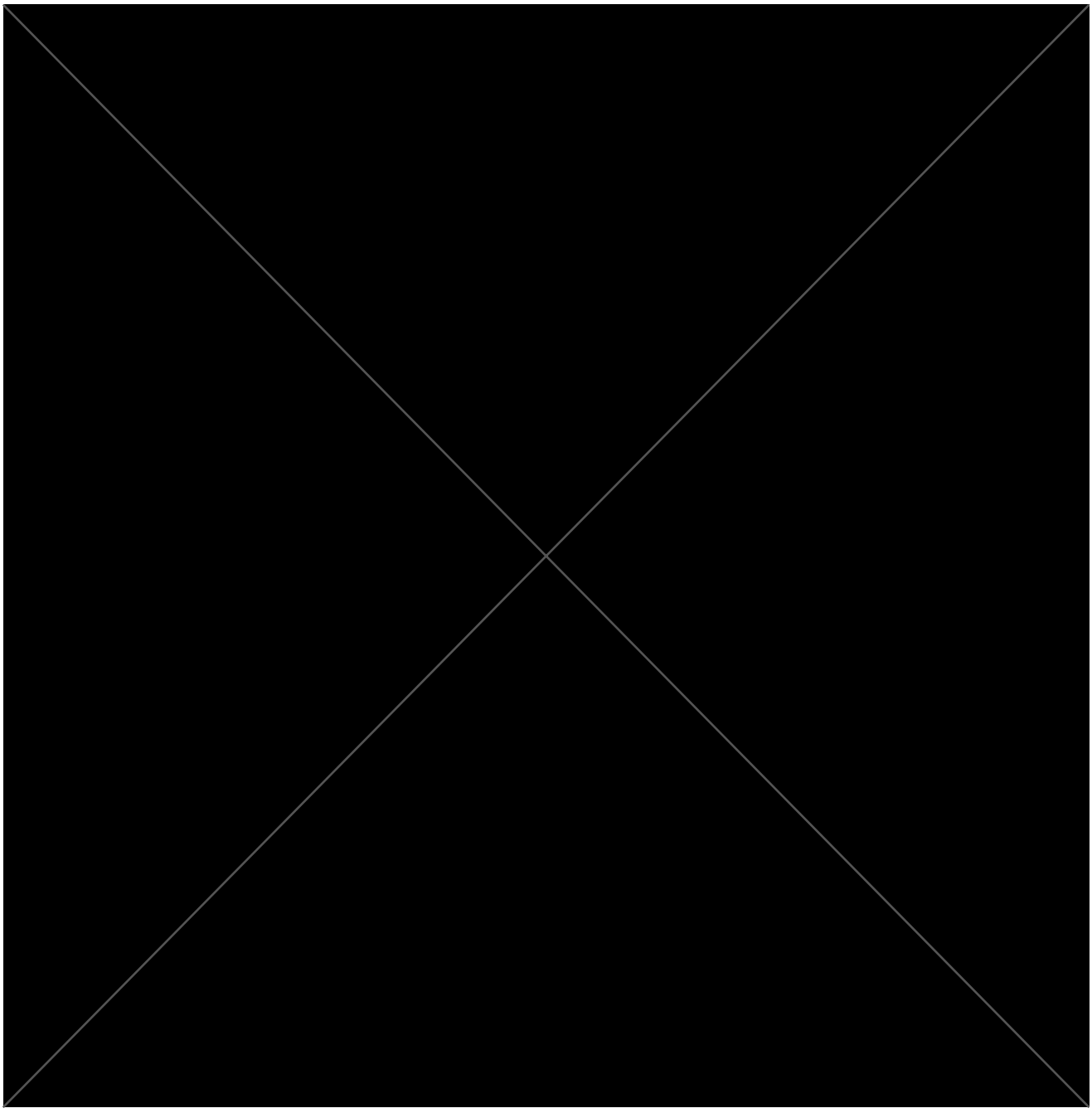

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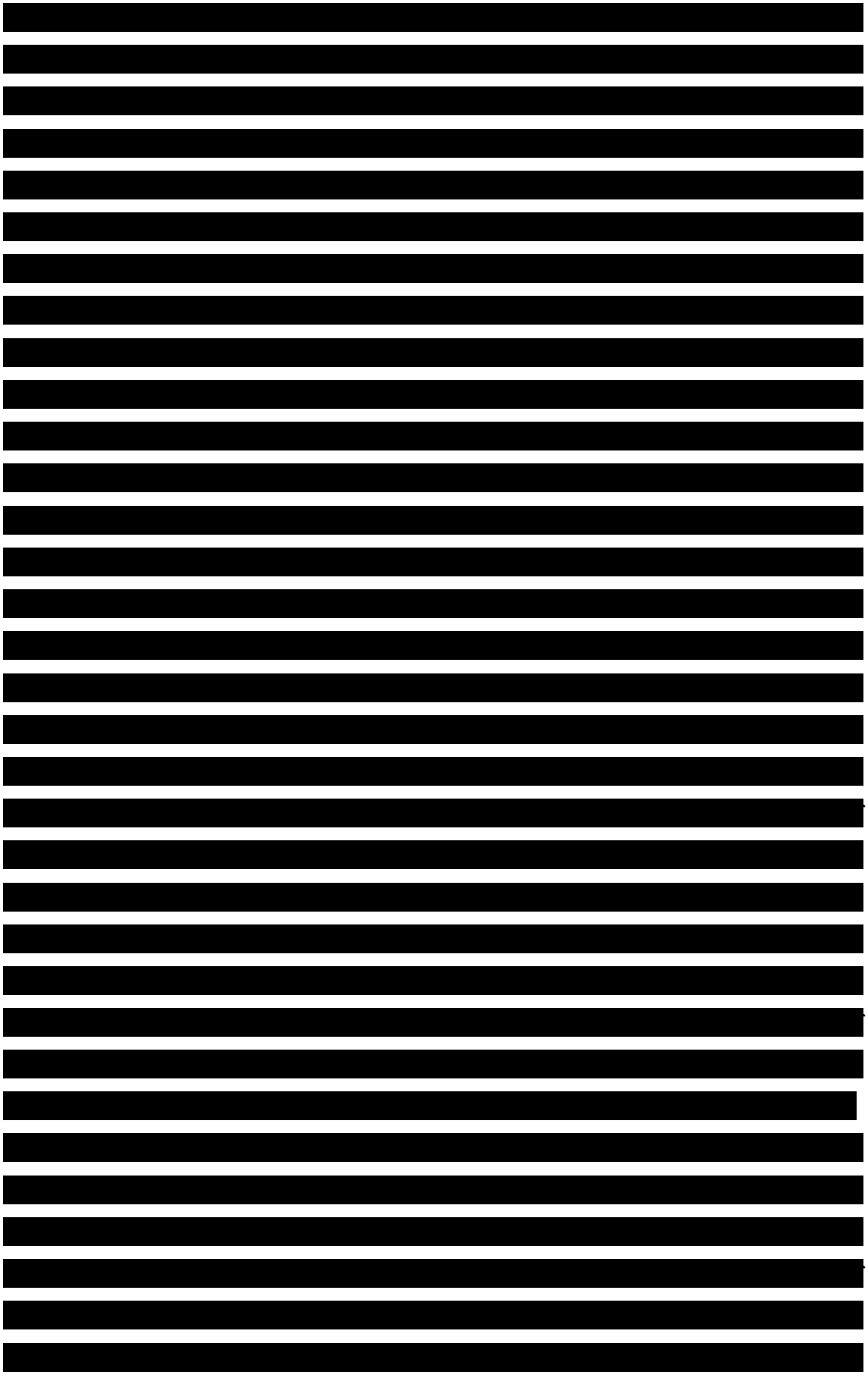

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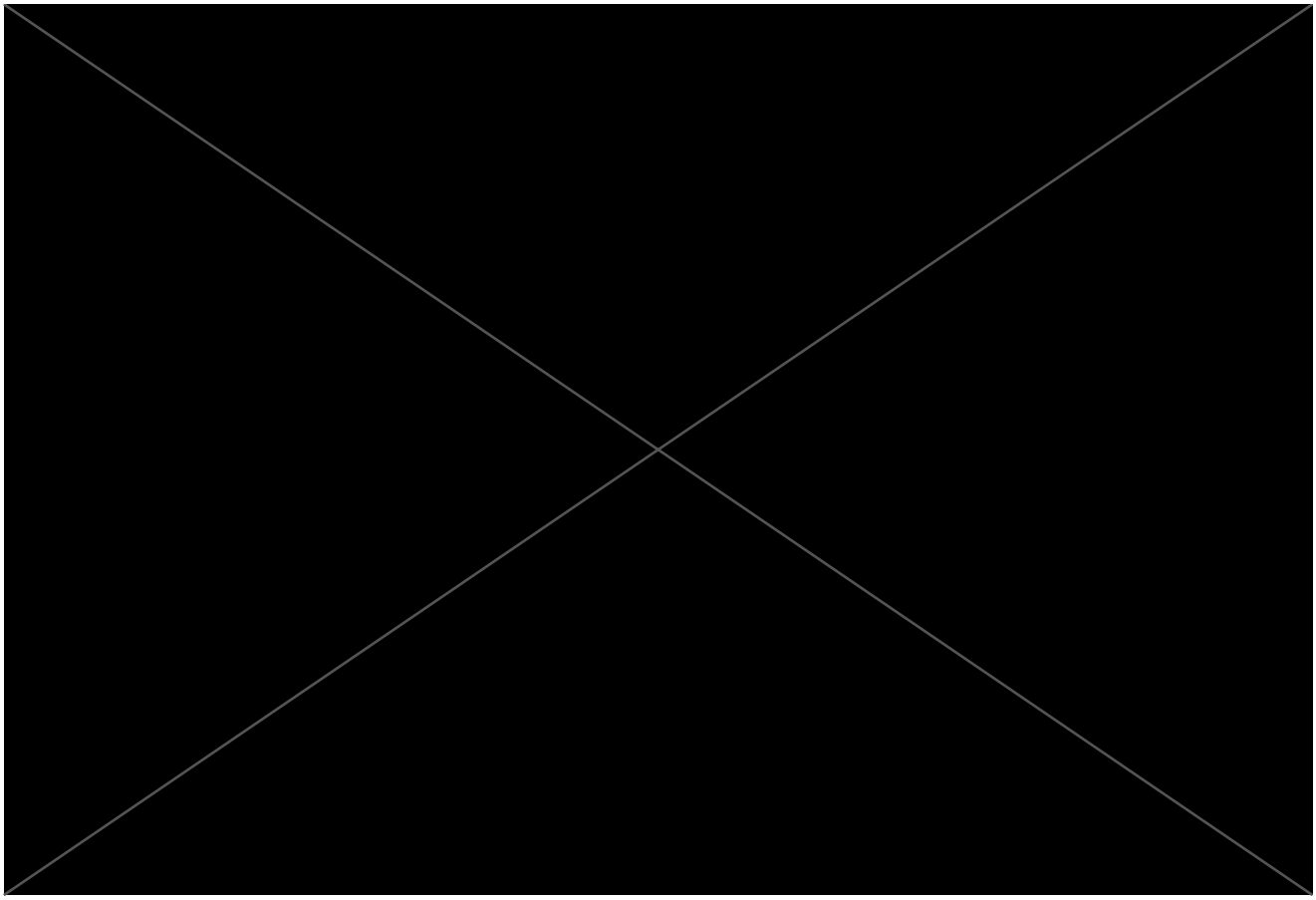

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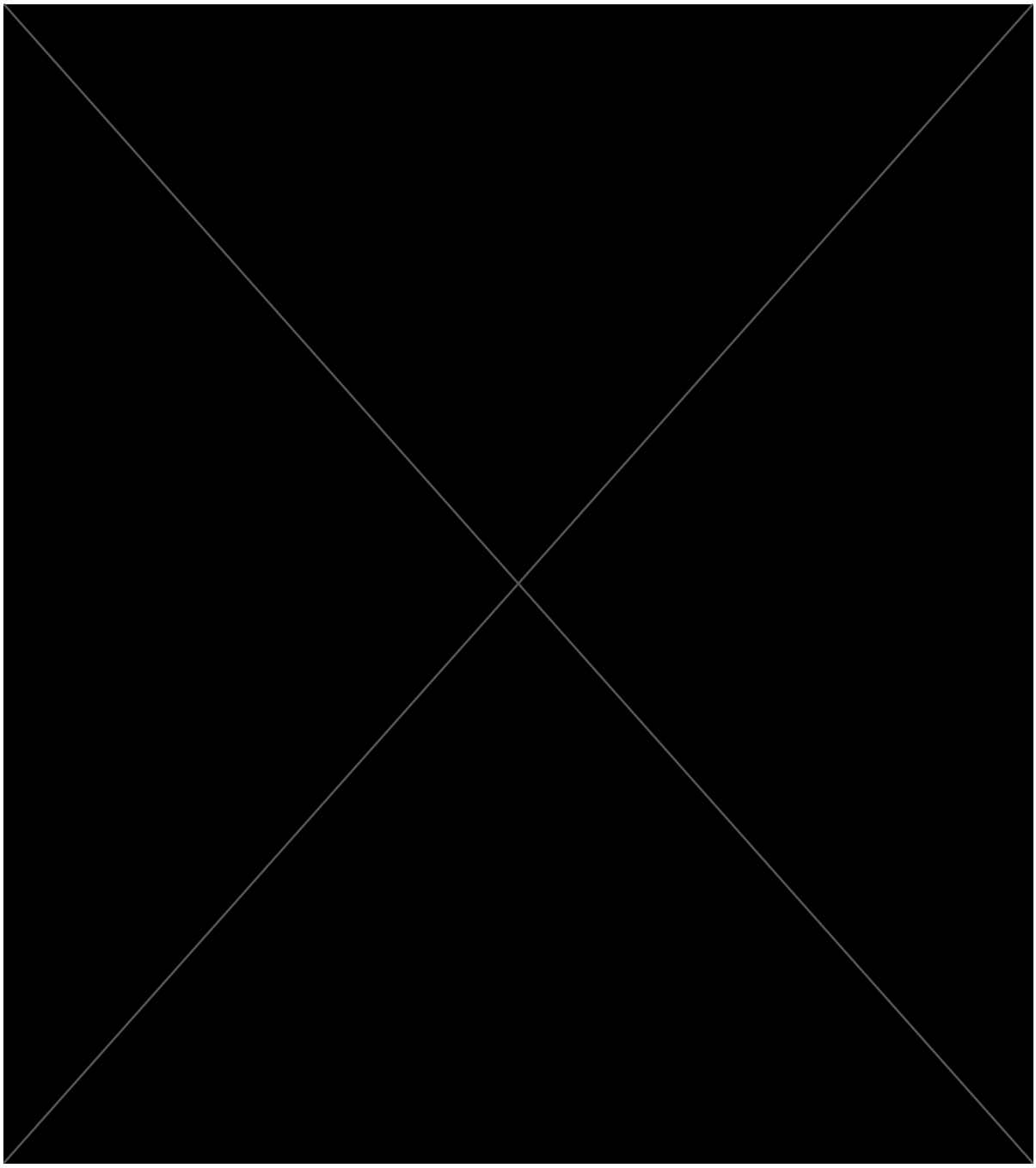

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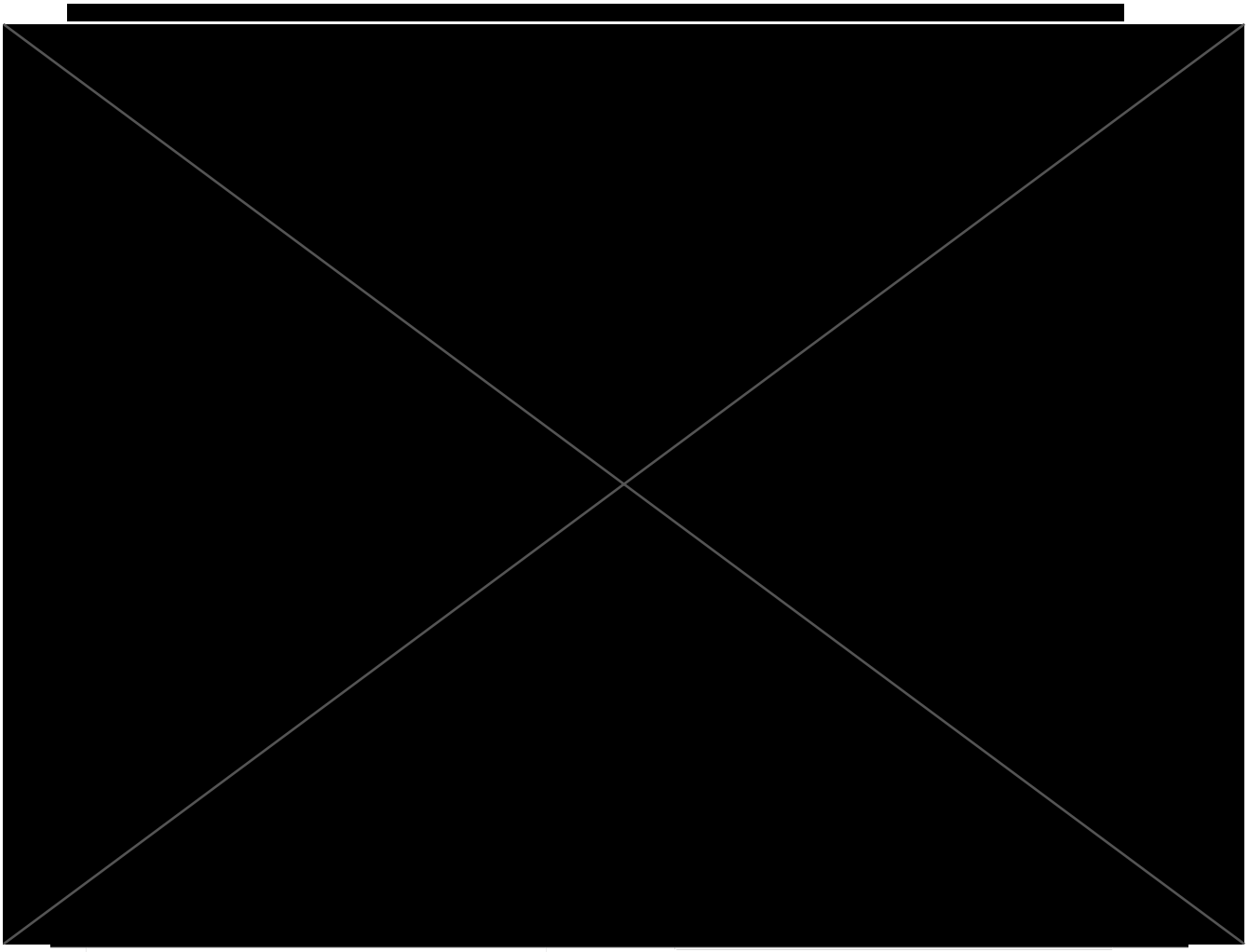

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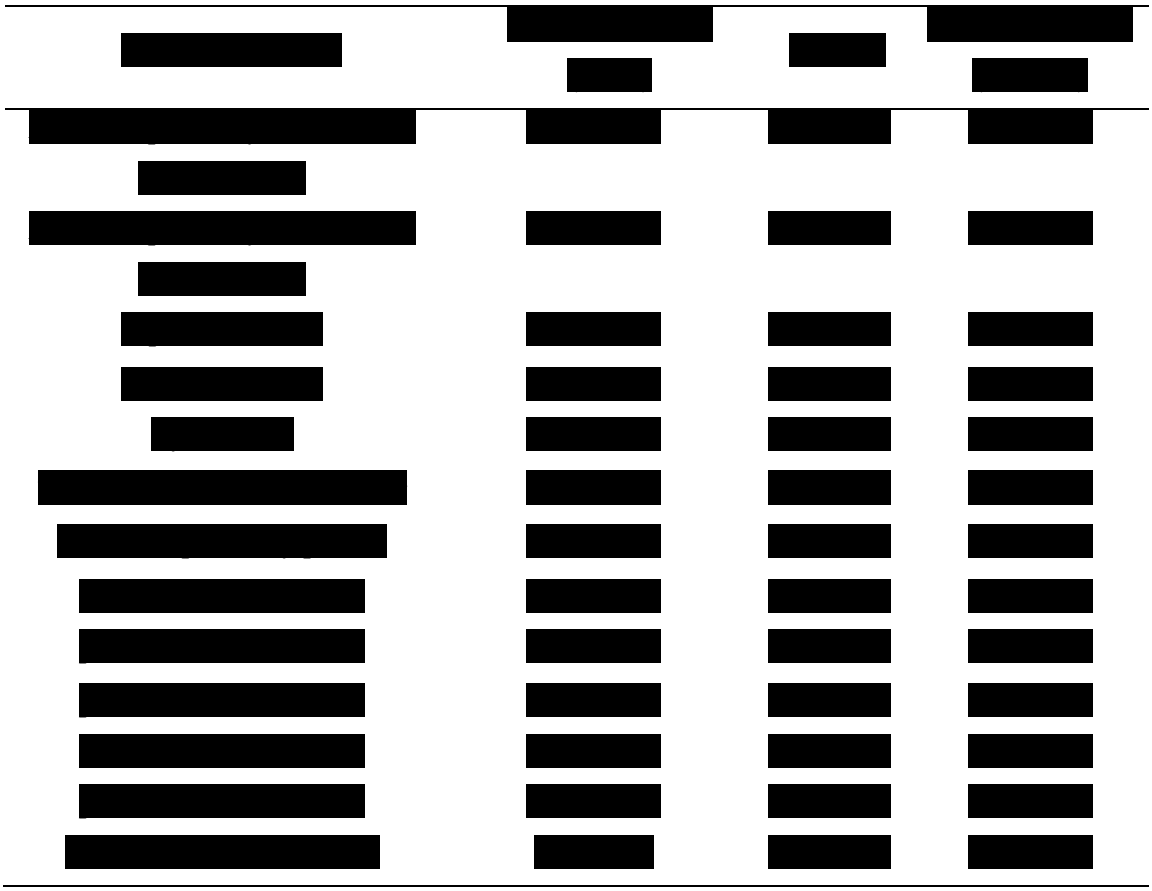

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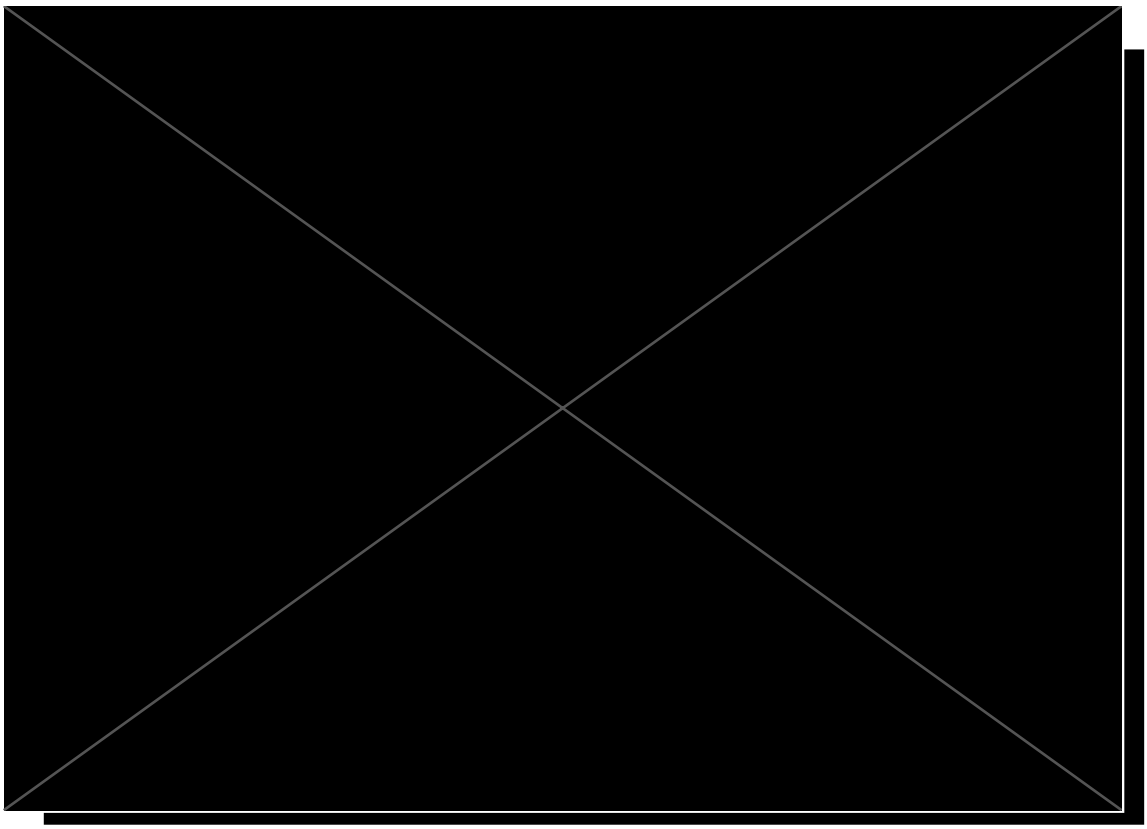

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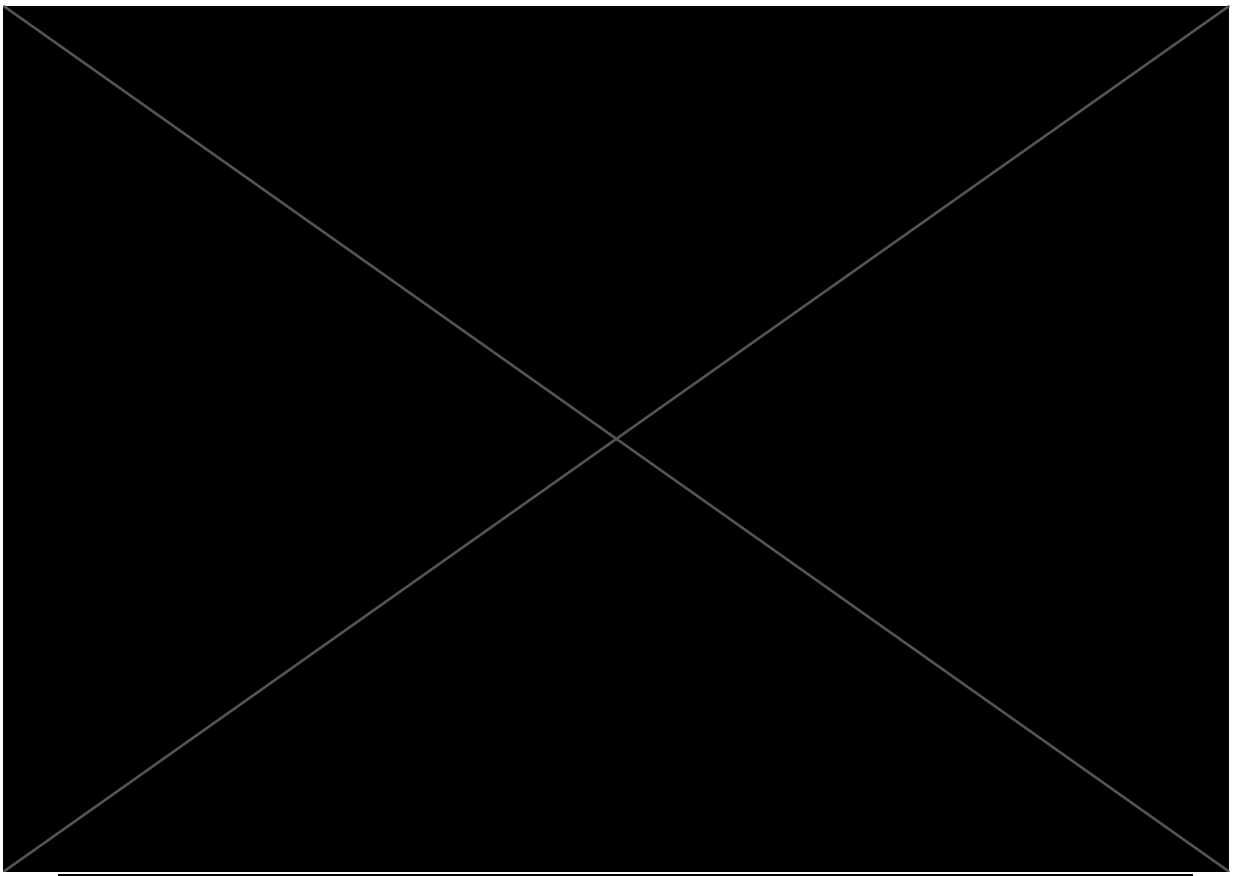

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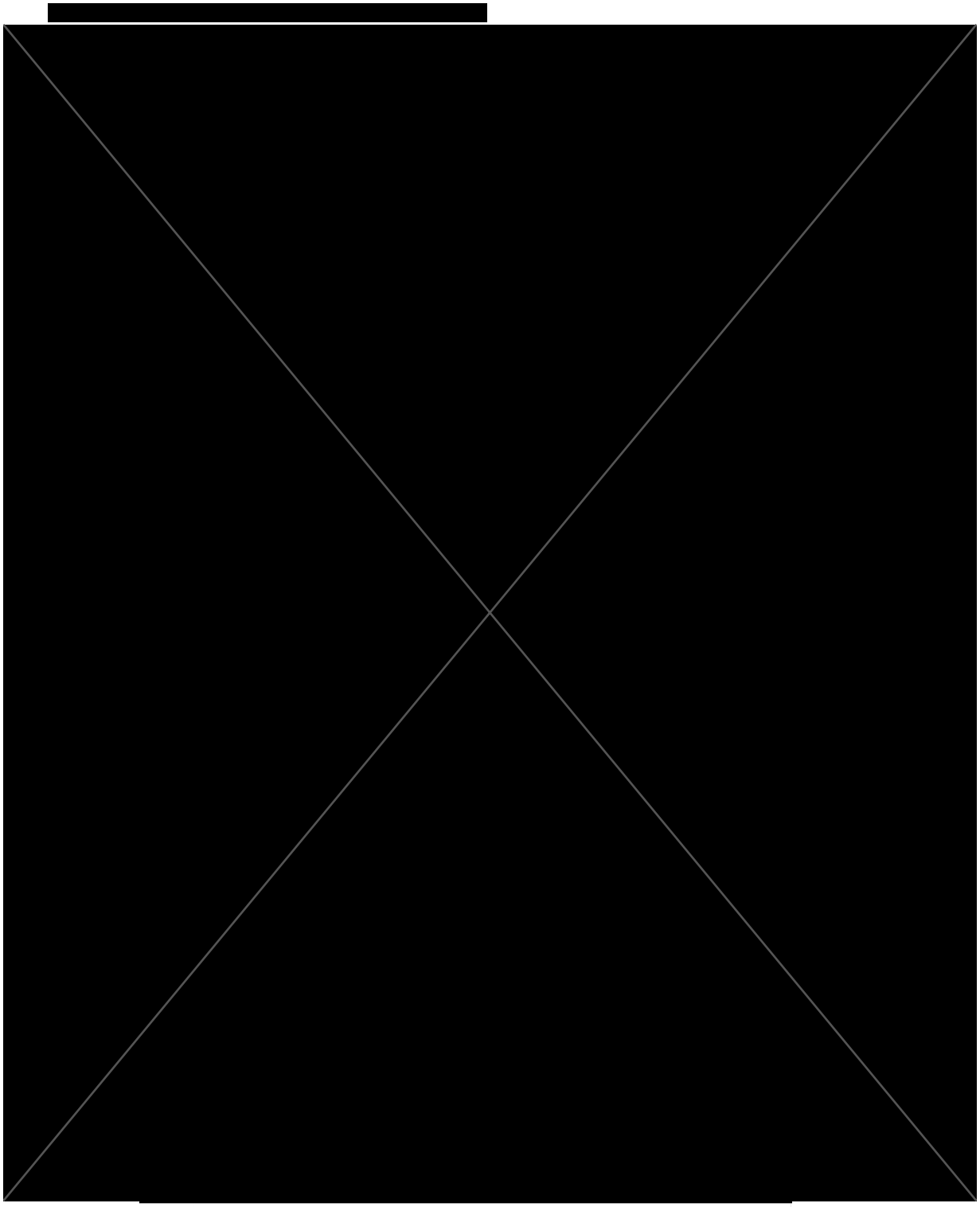

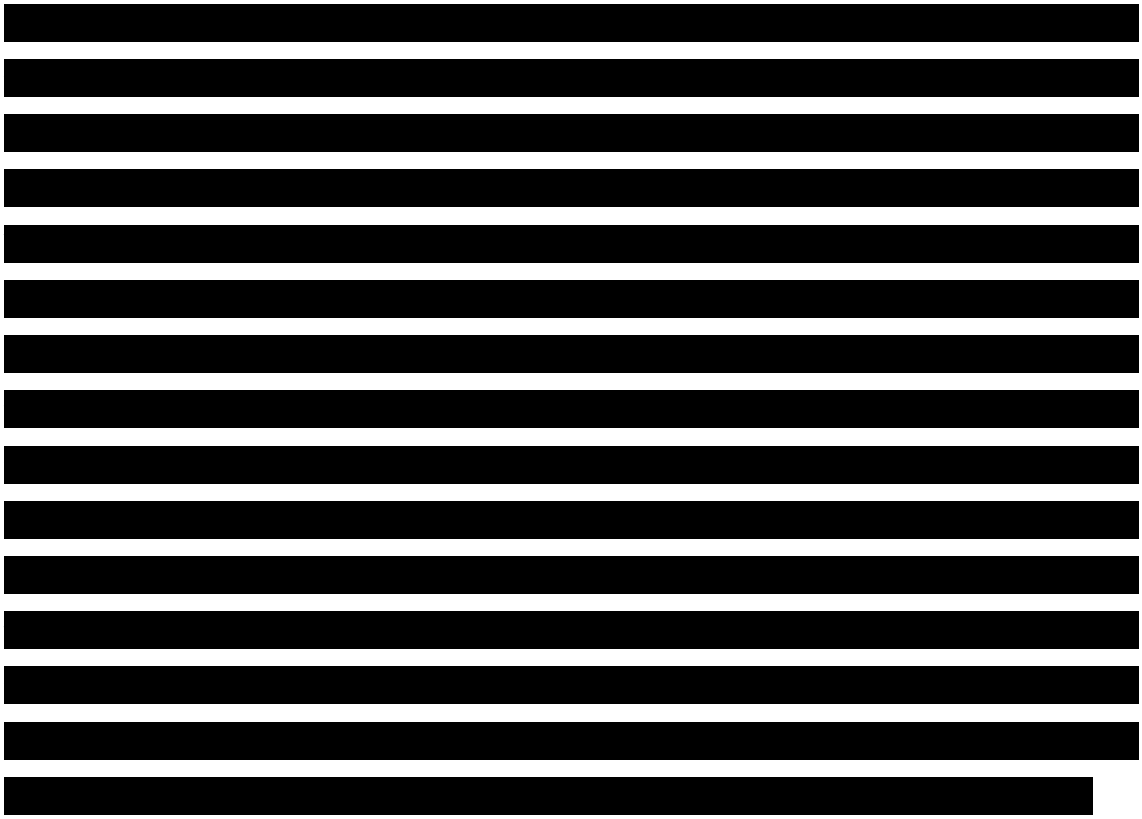

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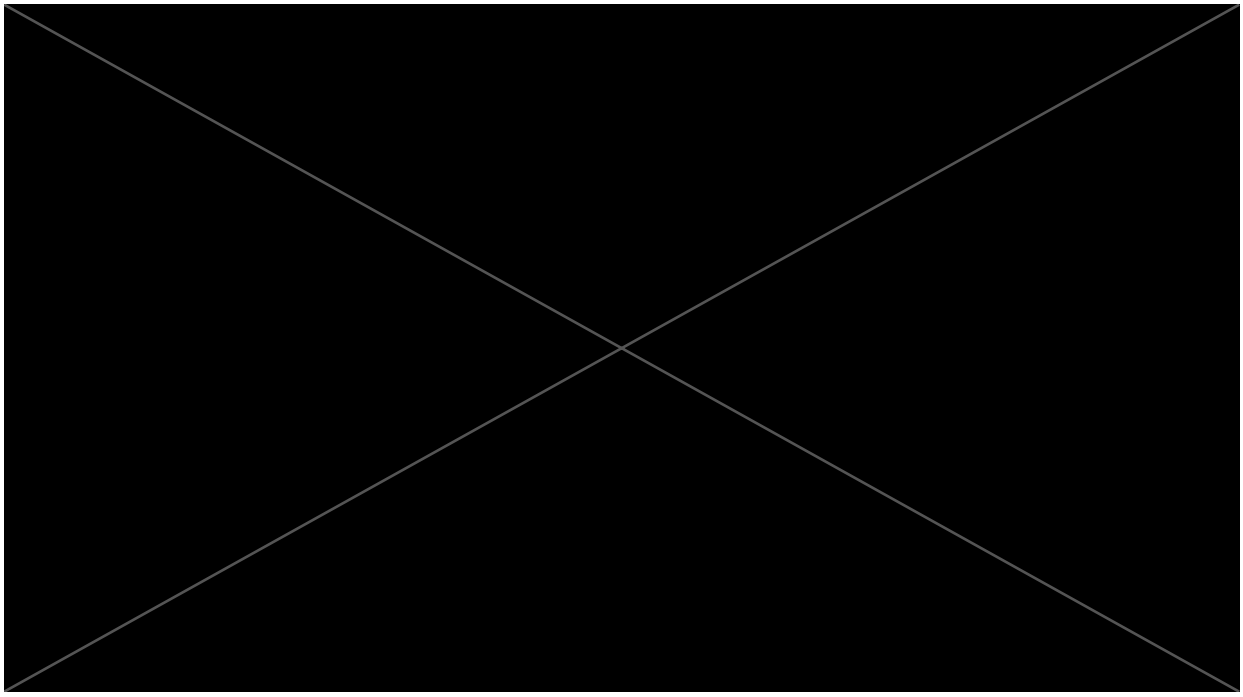

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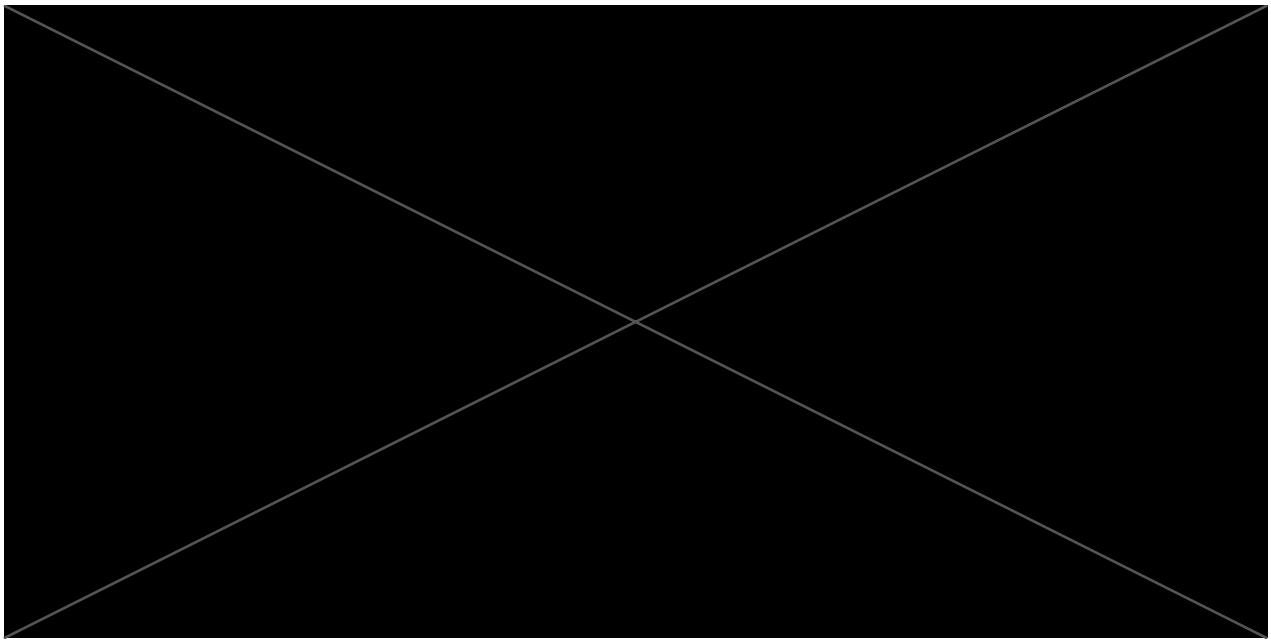

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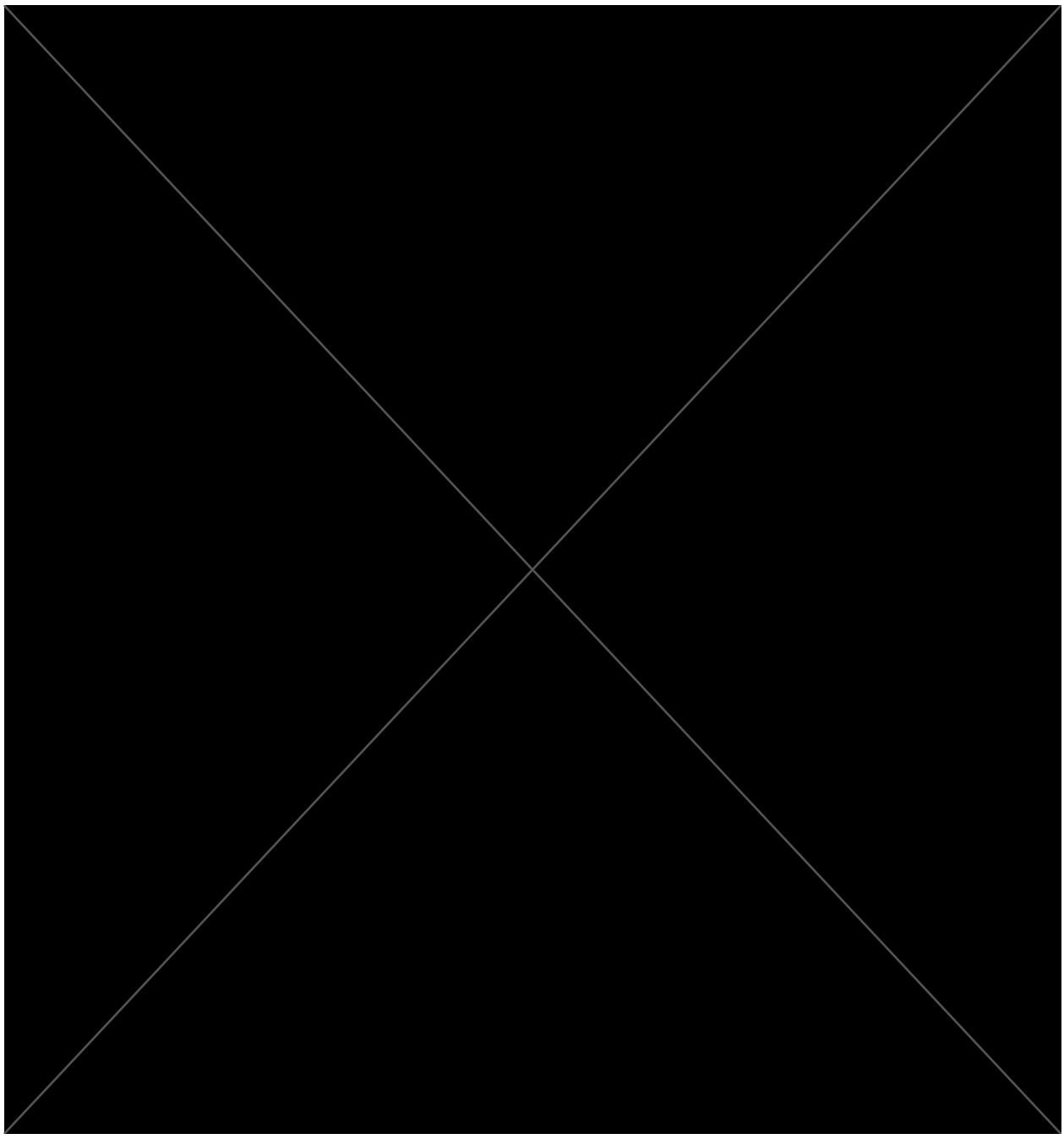

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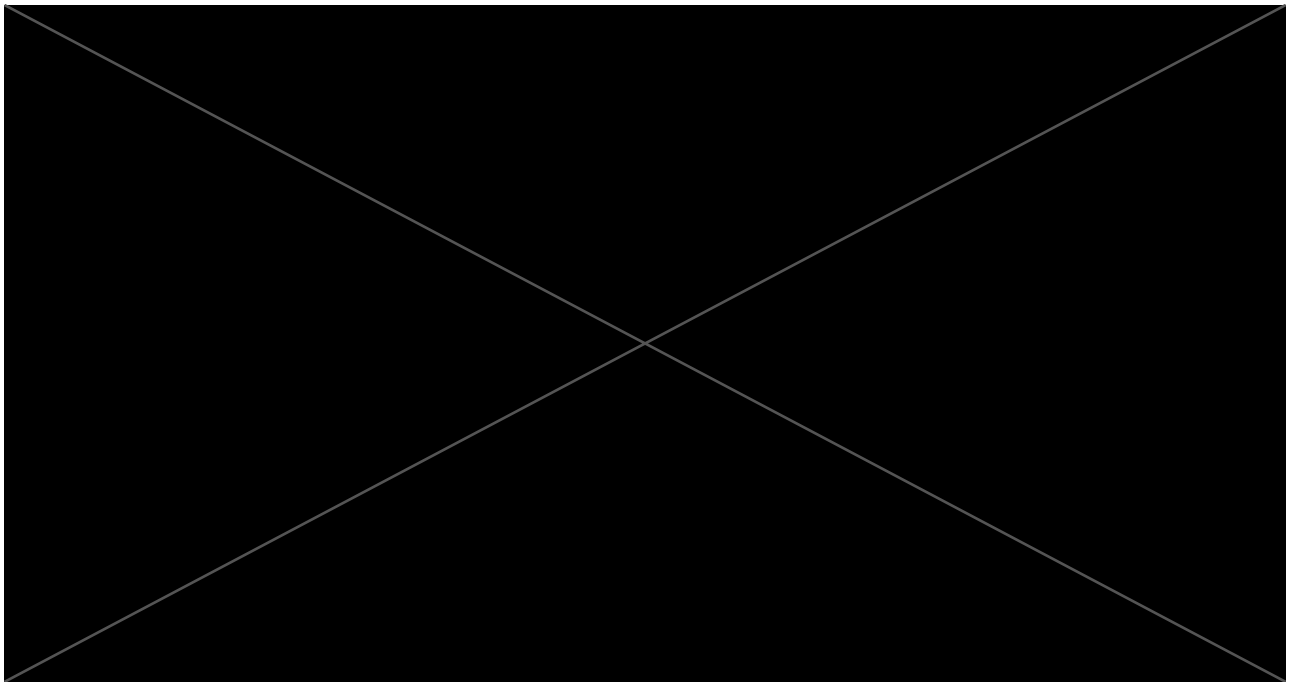

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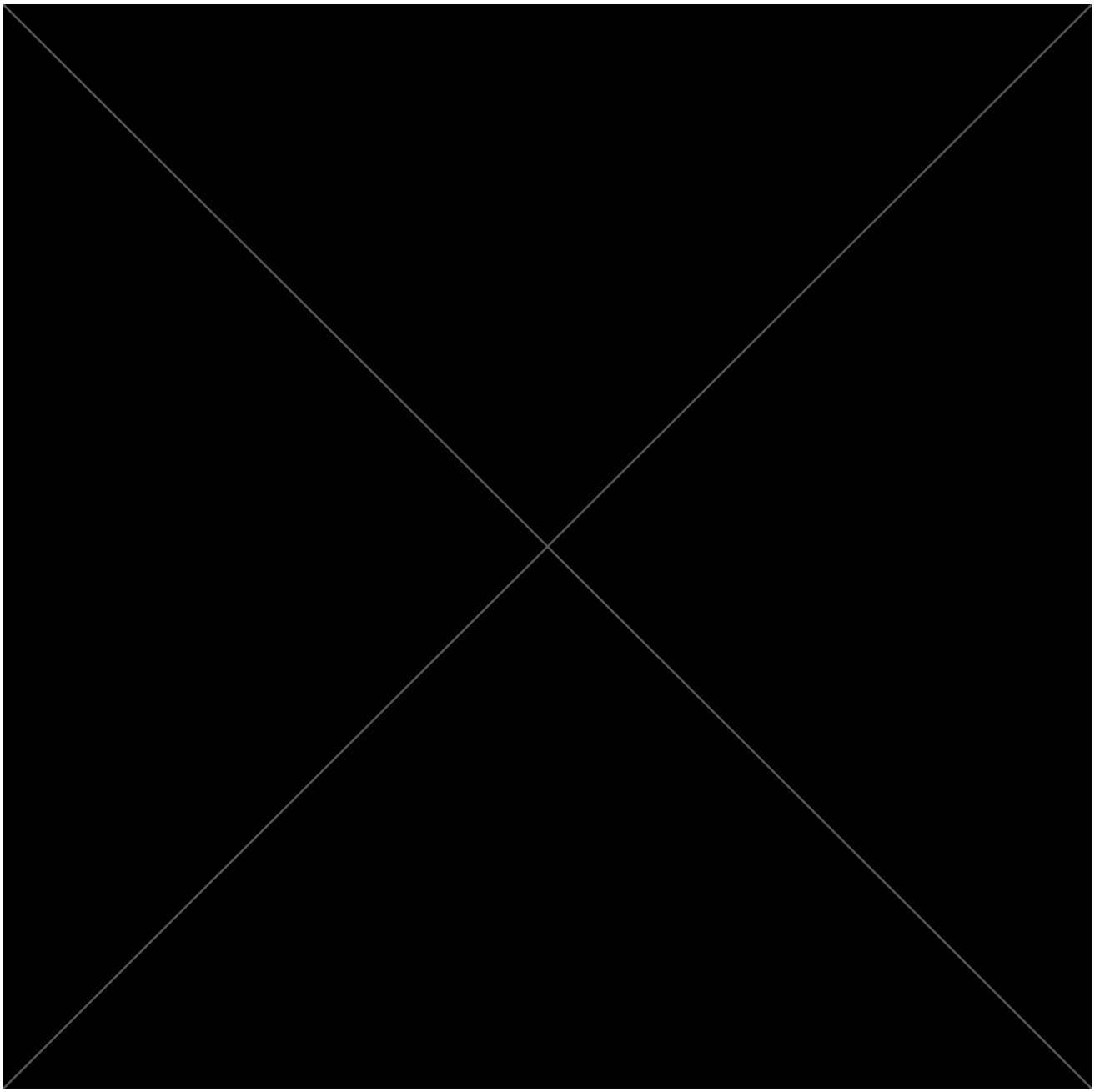

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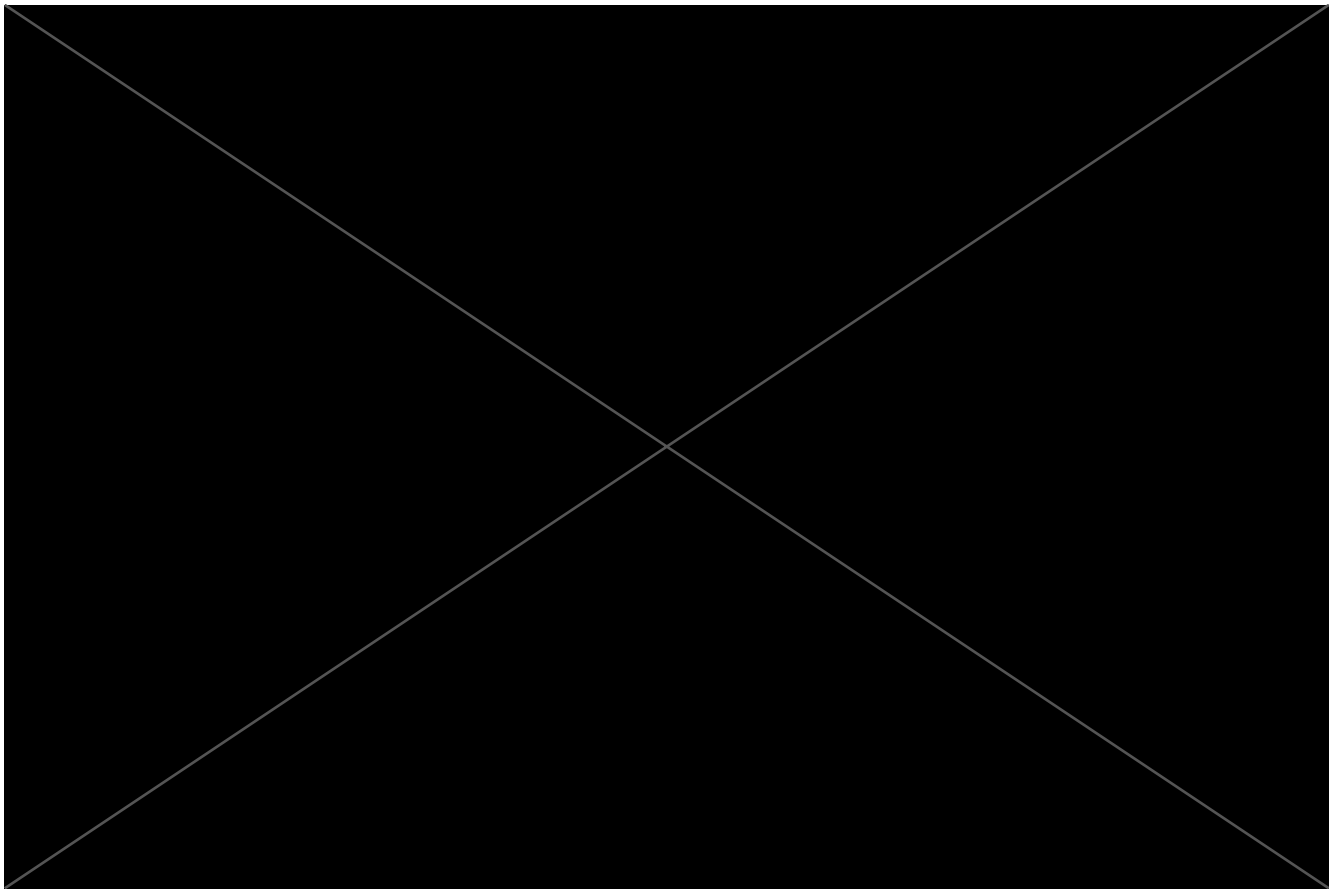

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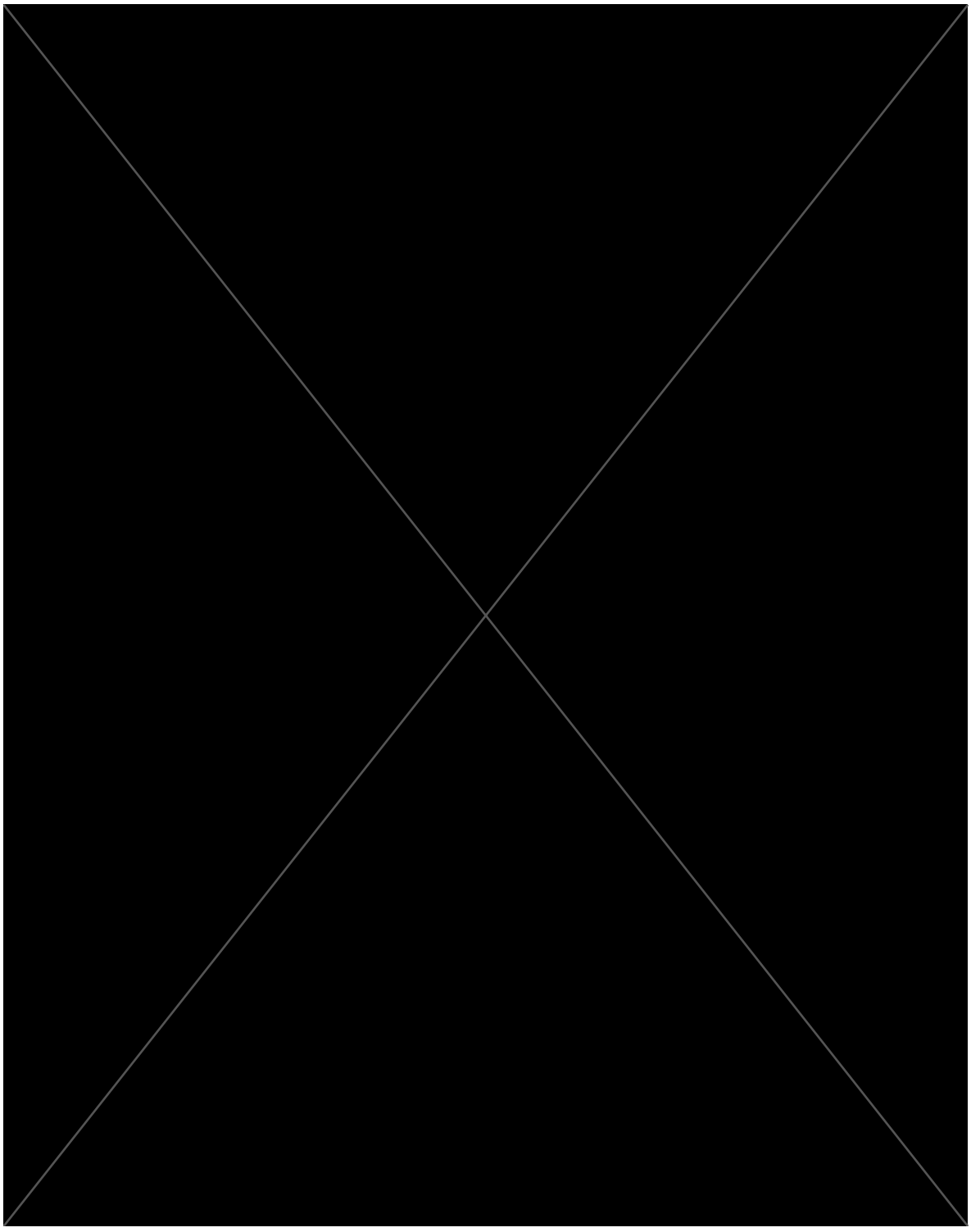

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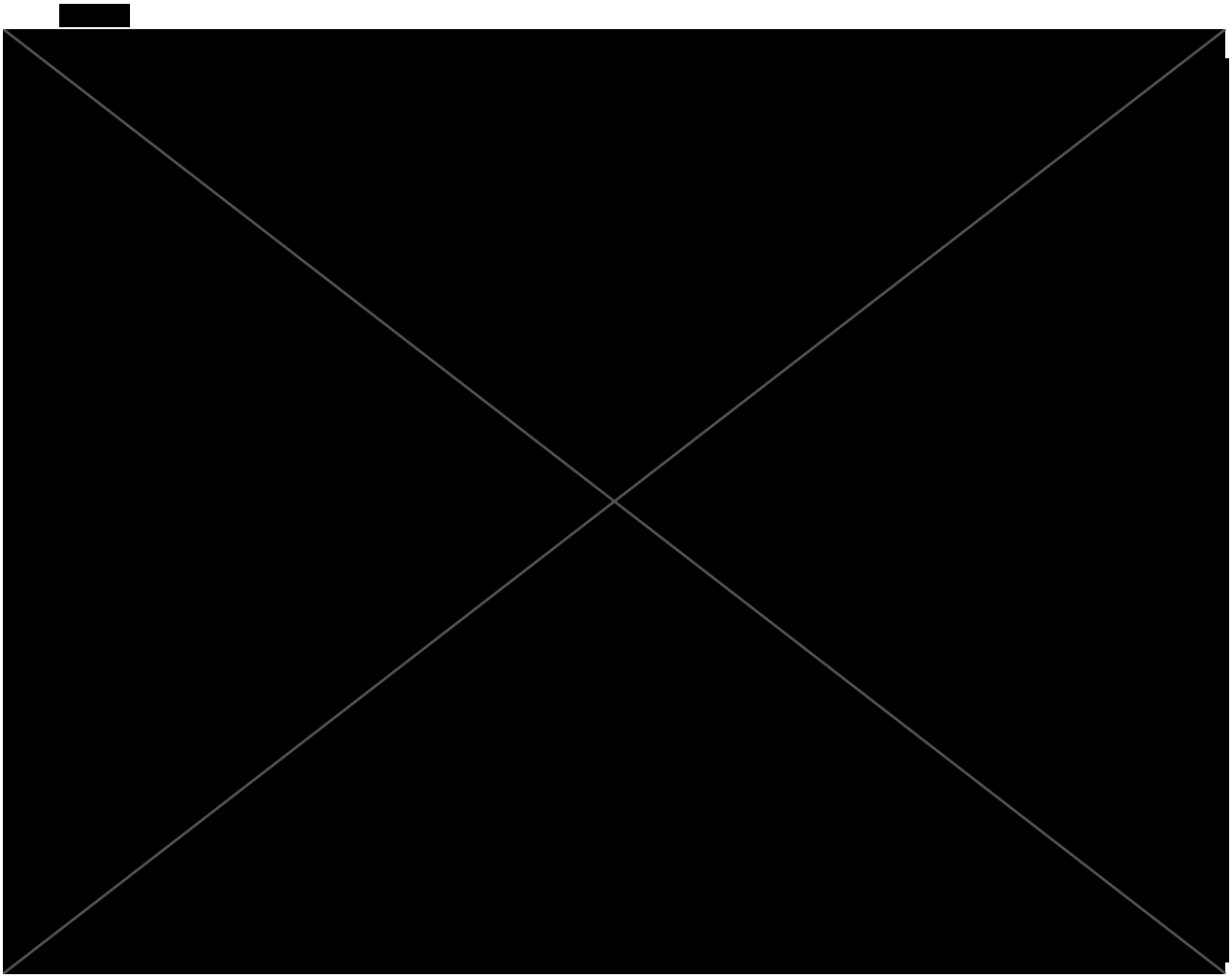

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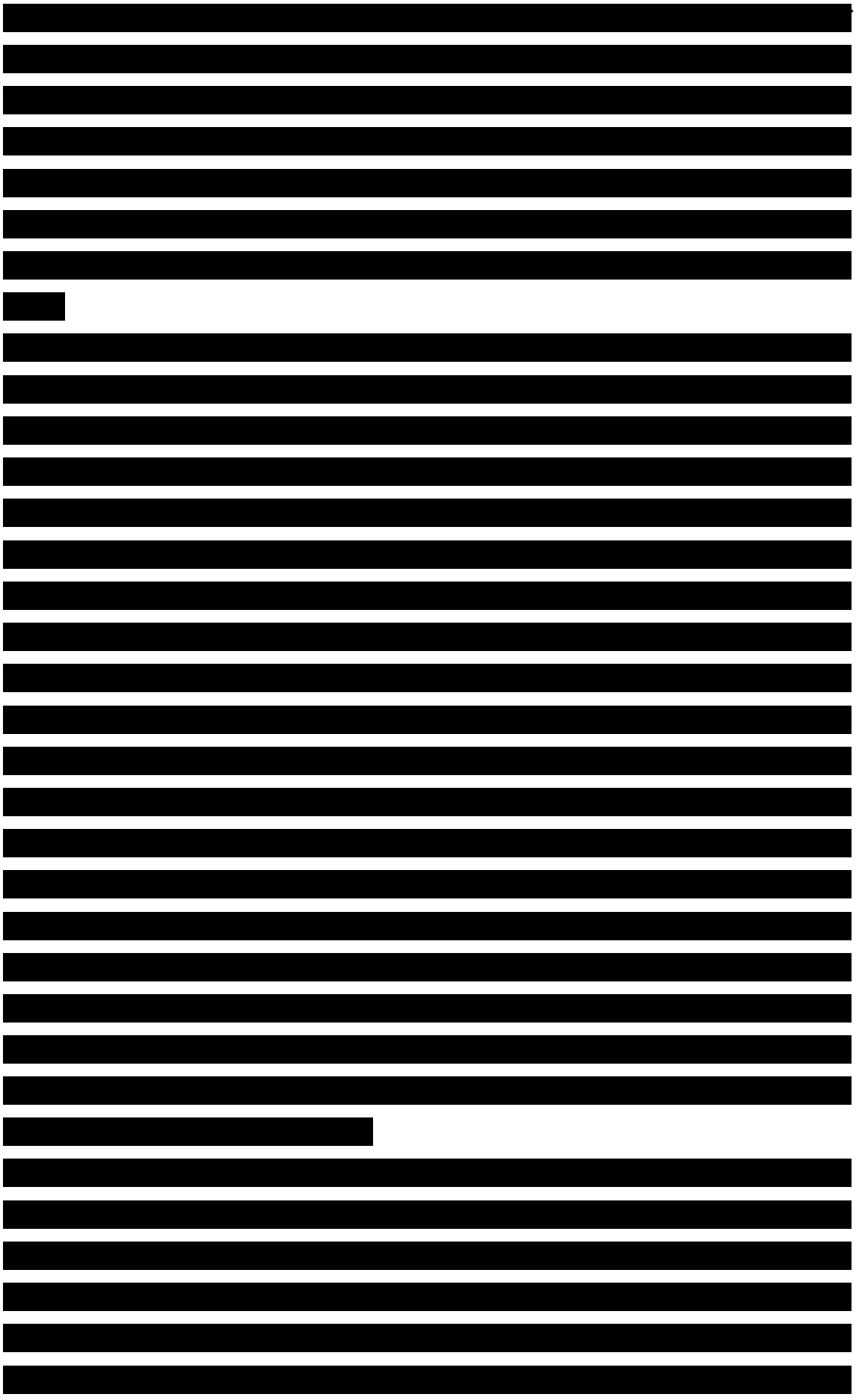

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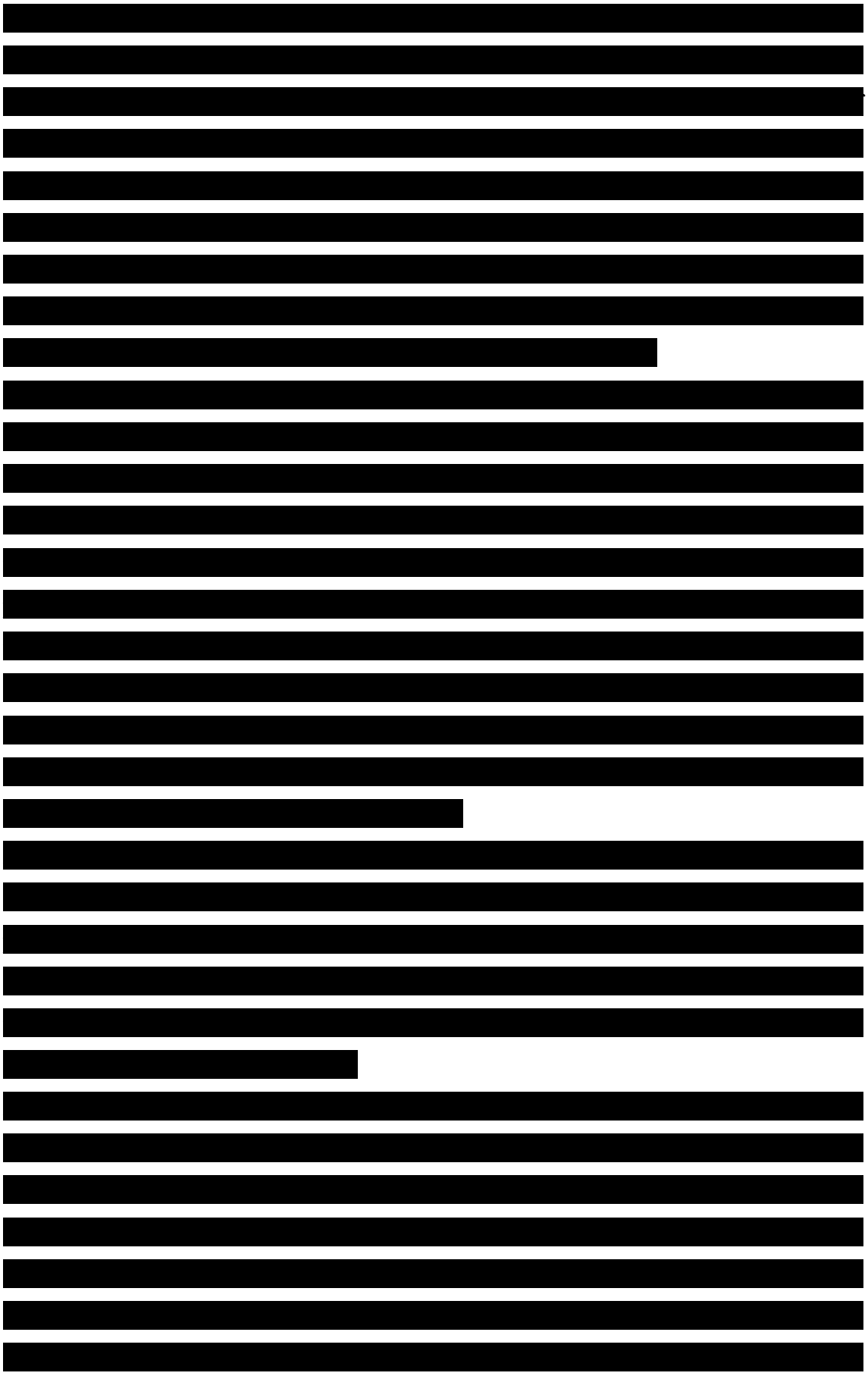

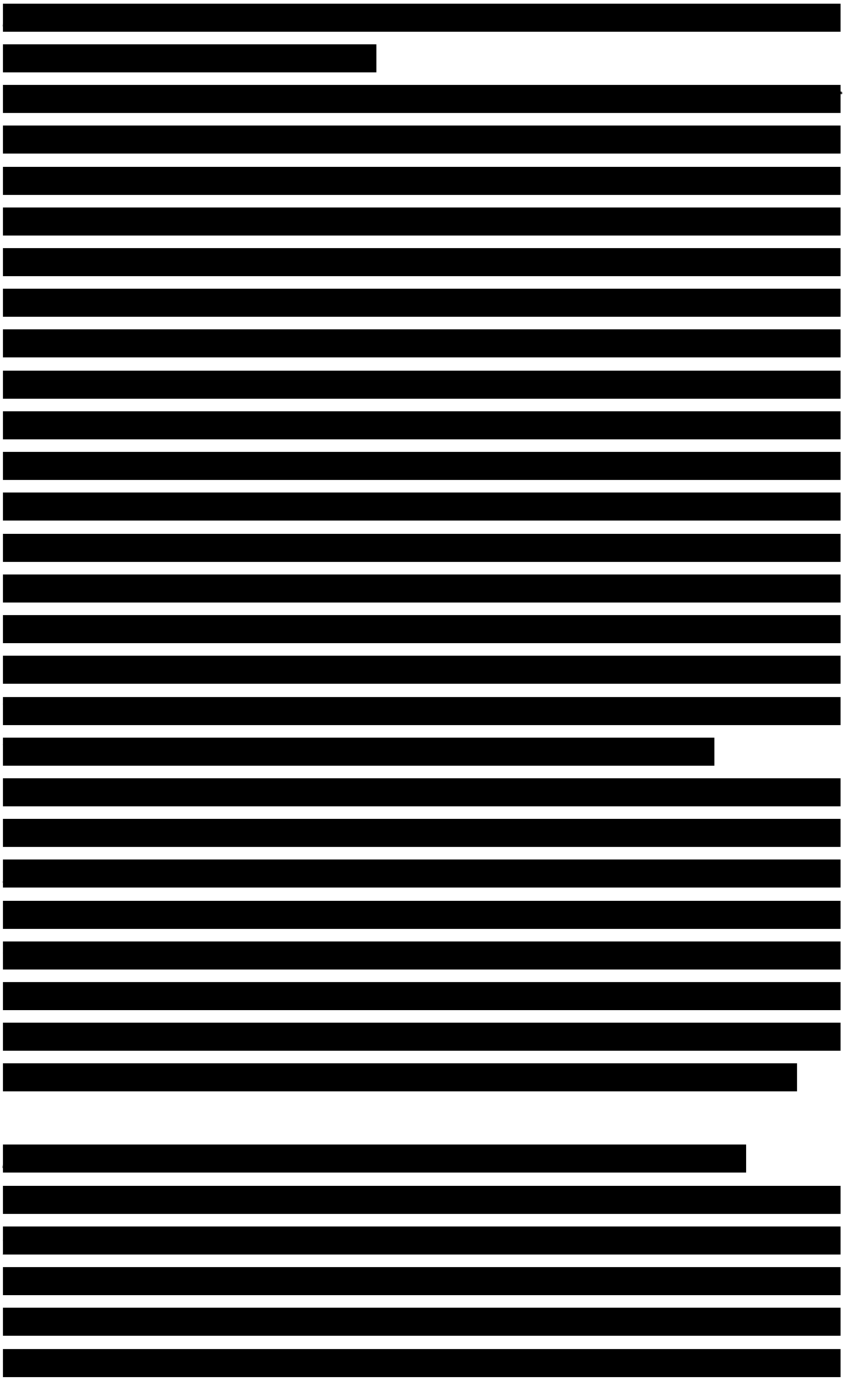

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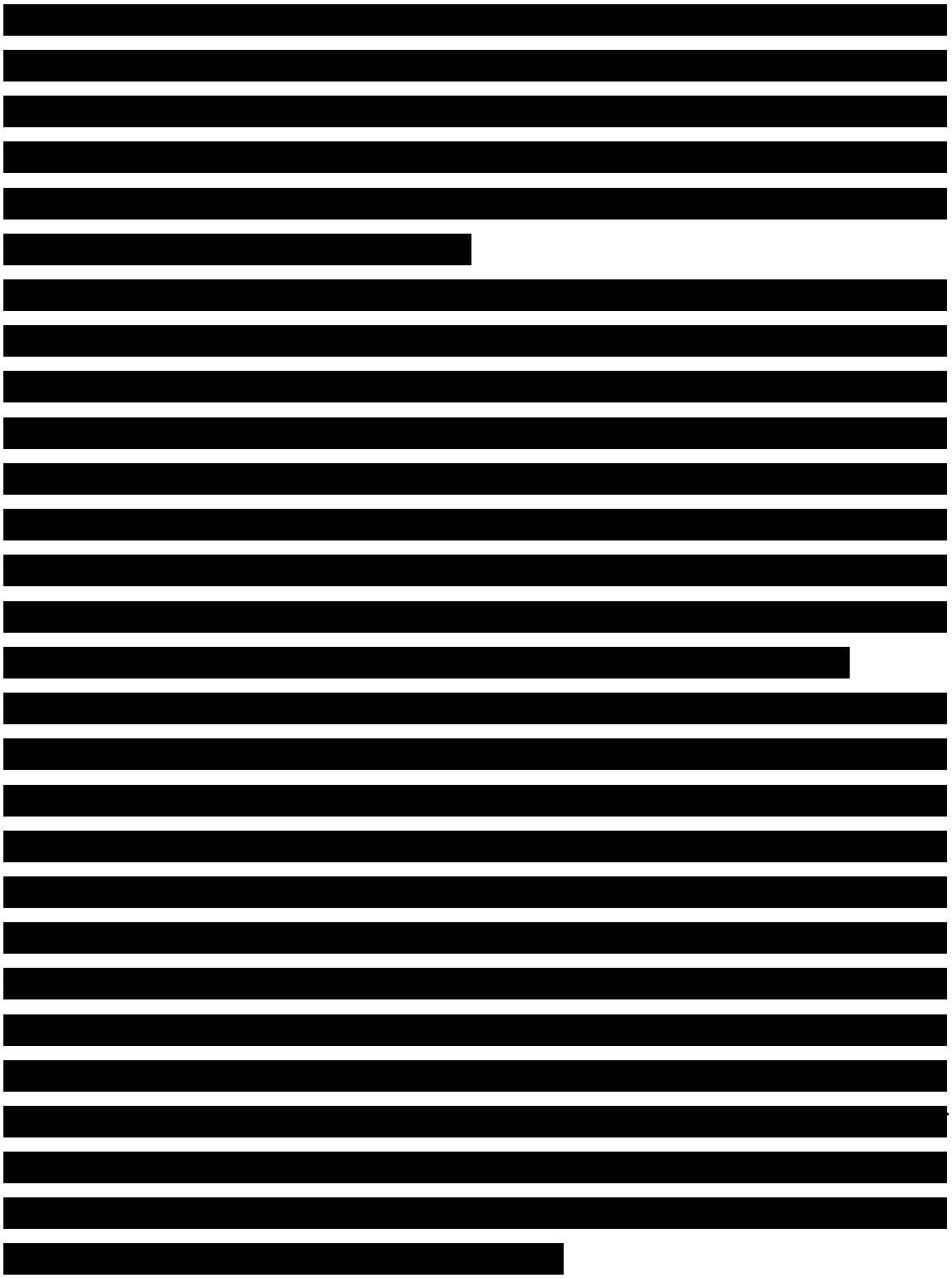

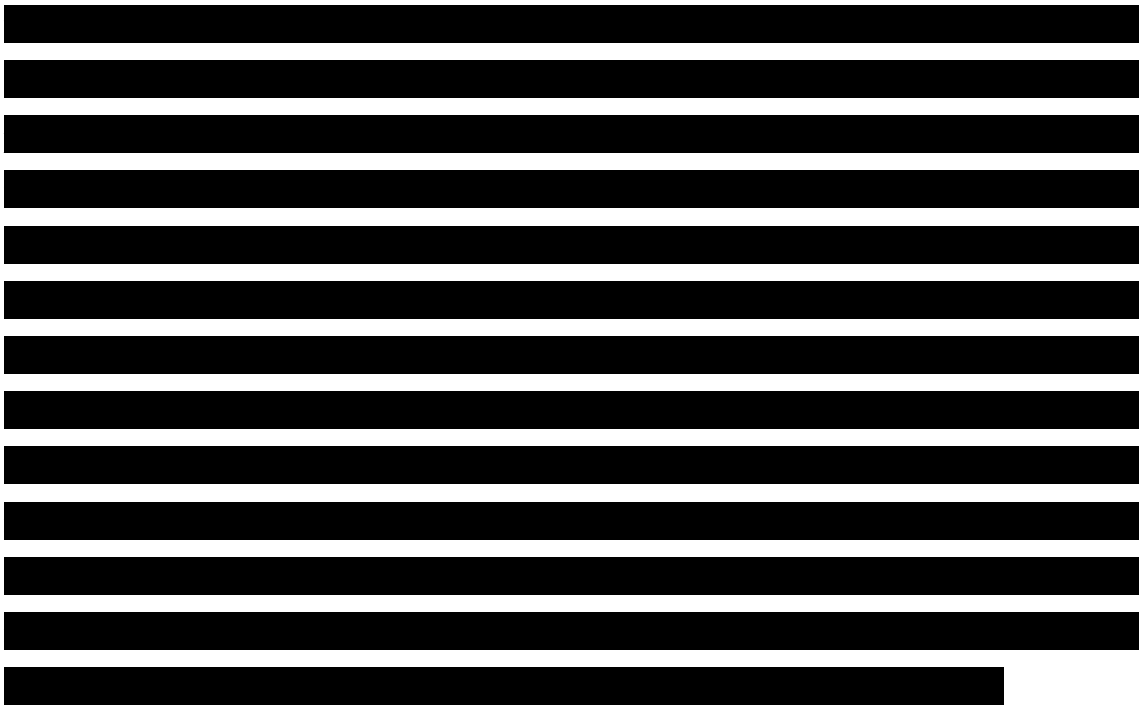

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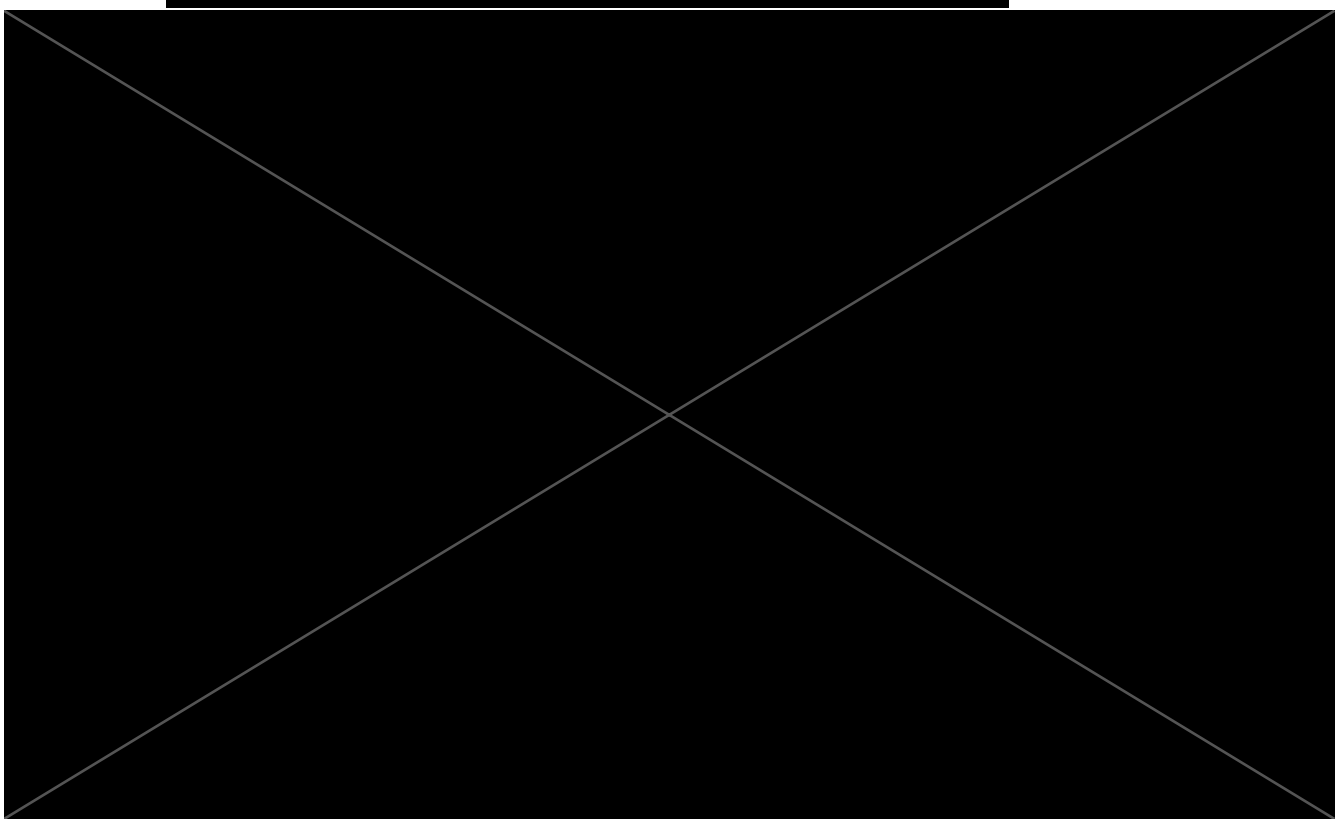

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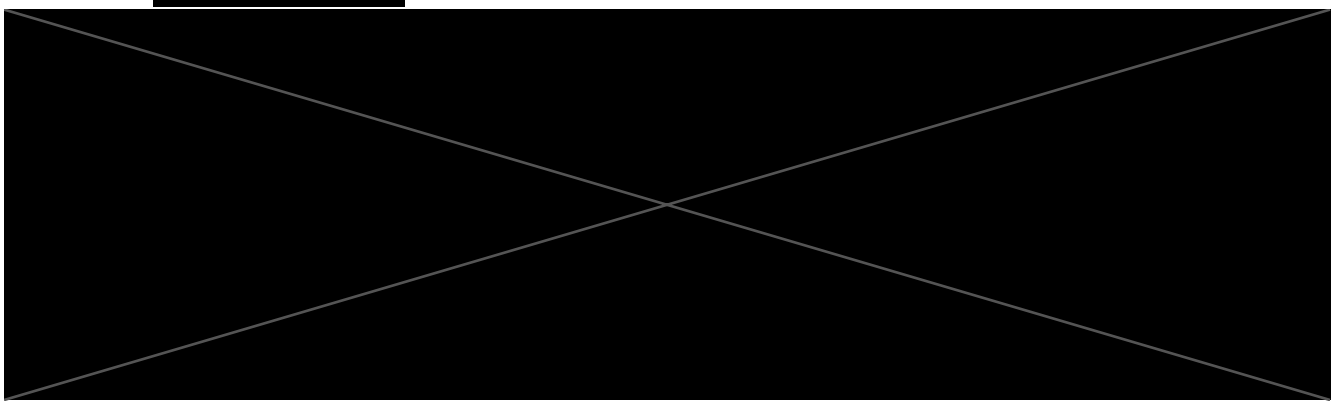
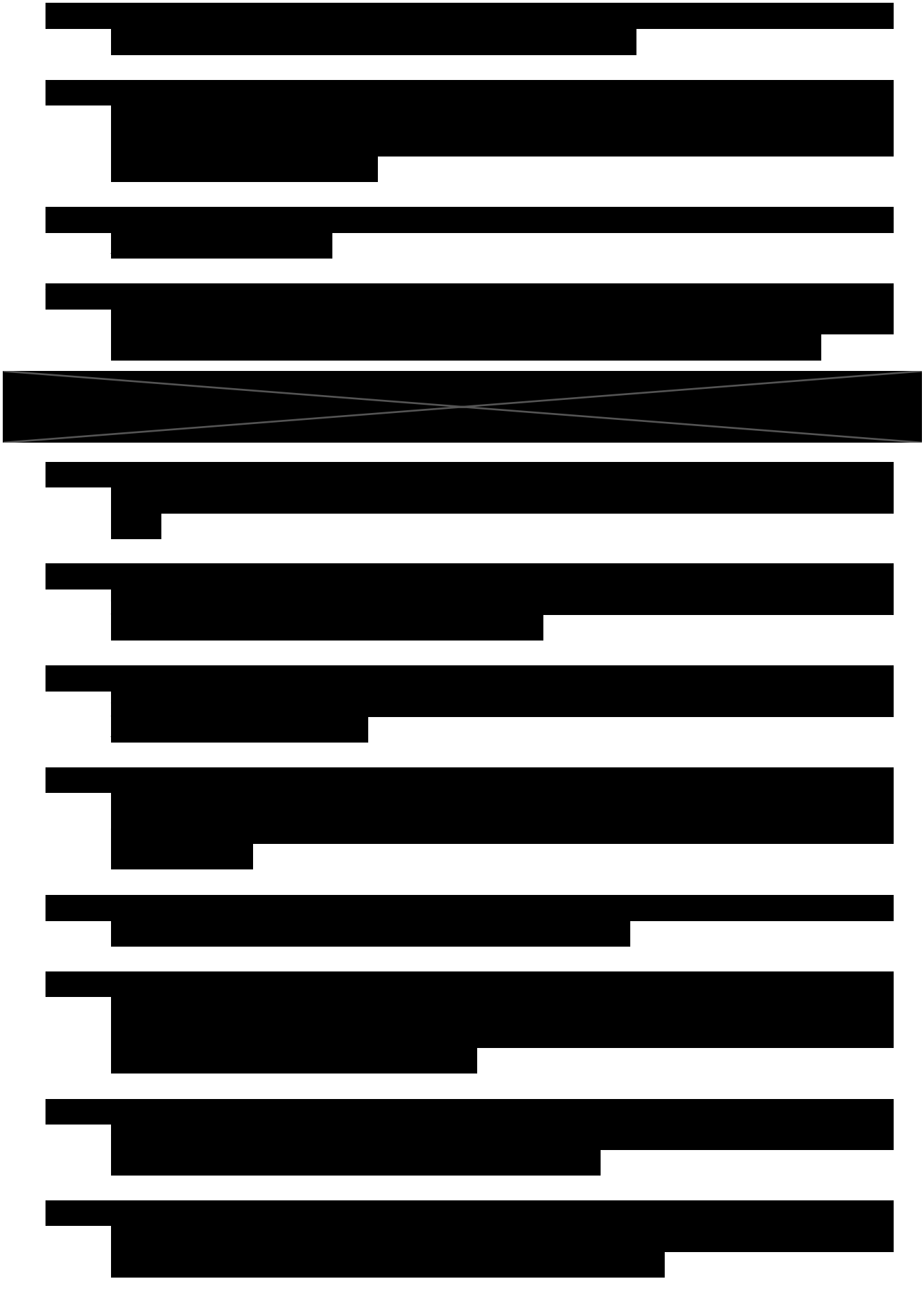
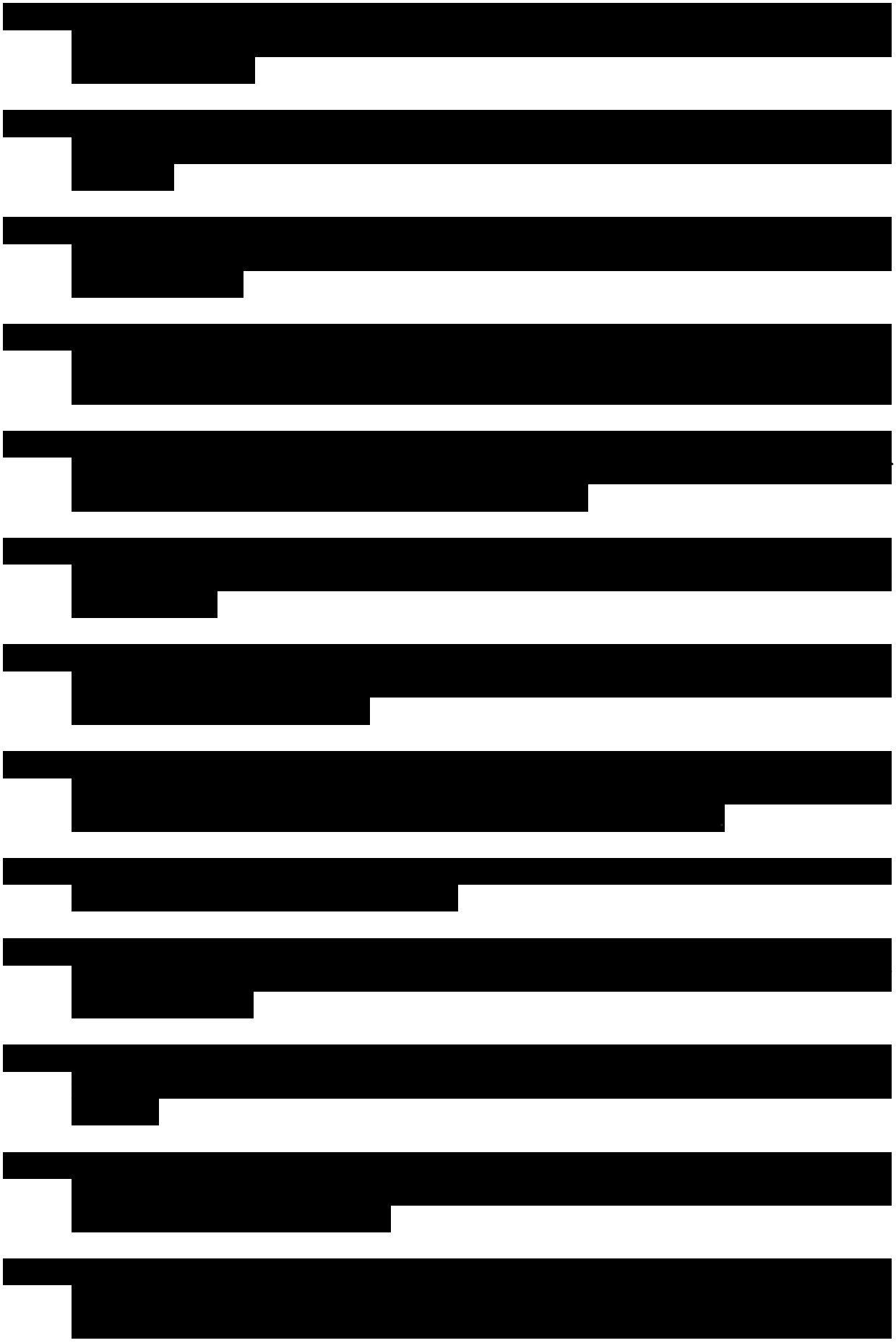
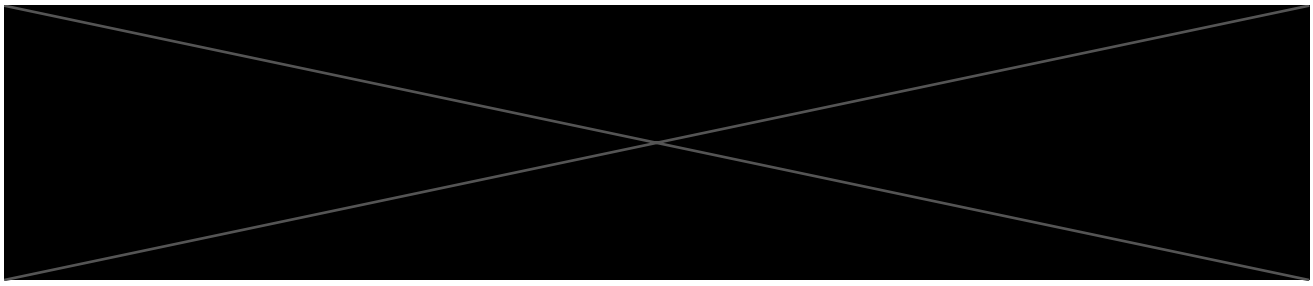
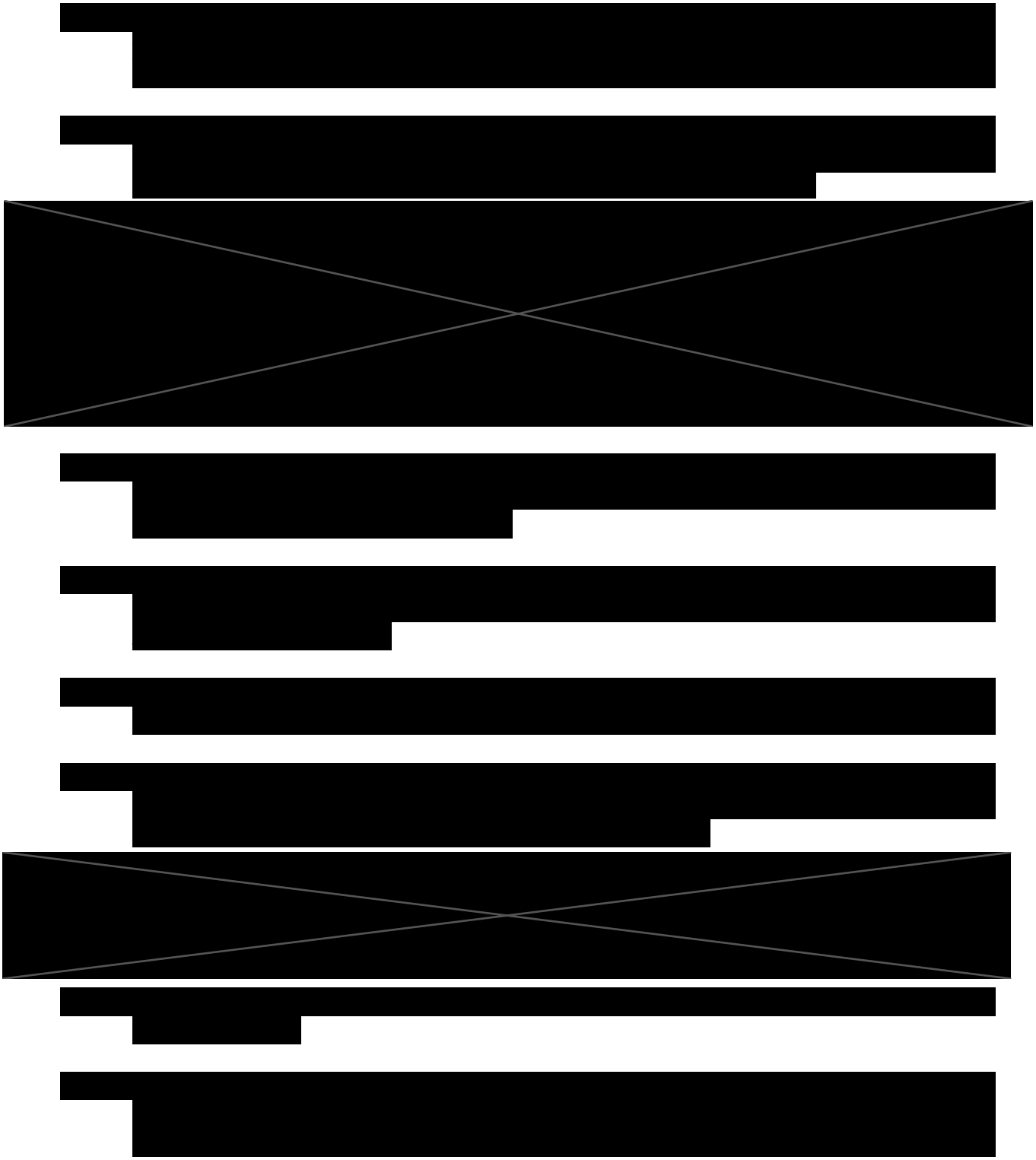
Choi, D., Lee, Y., Cho, H. T., & Kende, H. (2003). Regulation of expansin gene expression affects growth and development in transgenic rice plants. *The Plant Cell*, *15*(6), 1386-1398.











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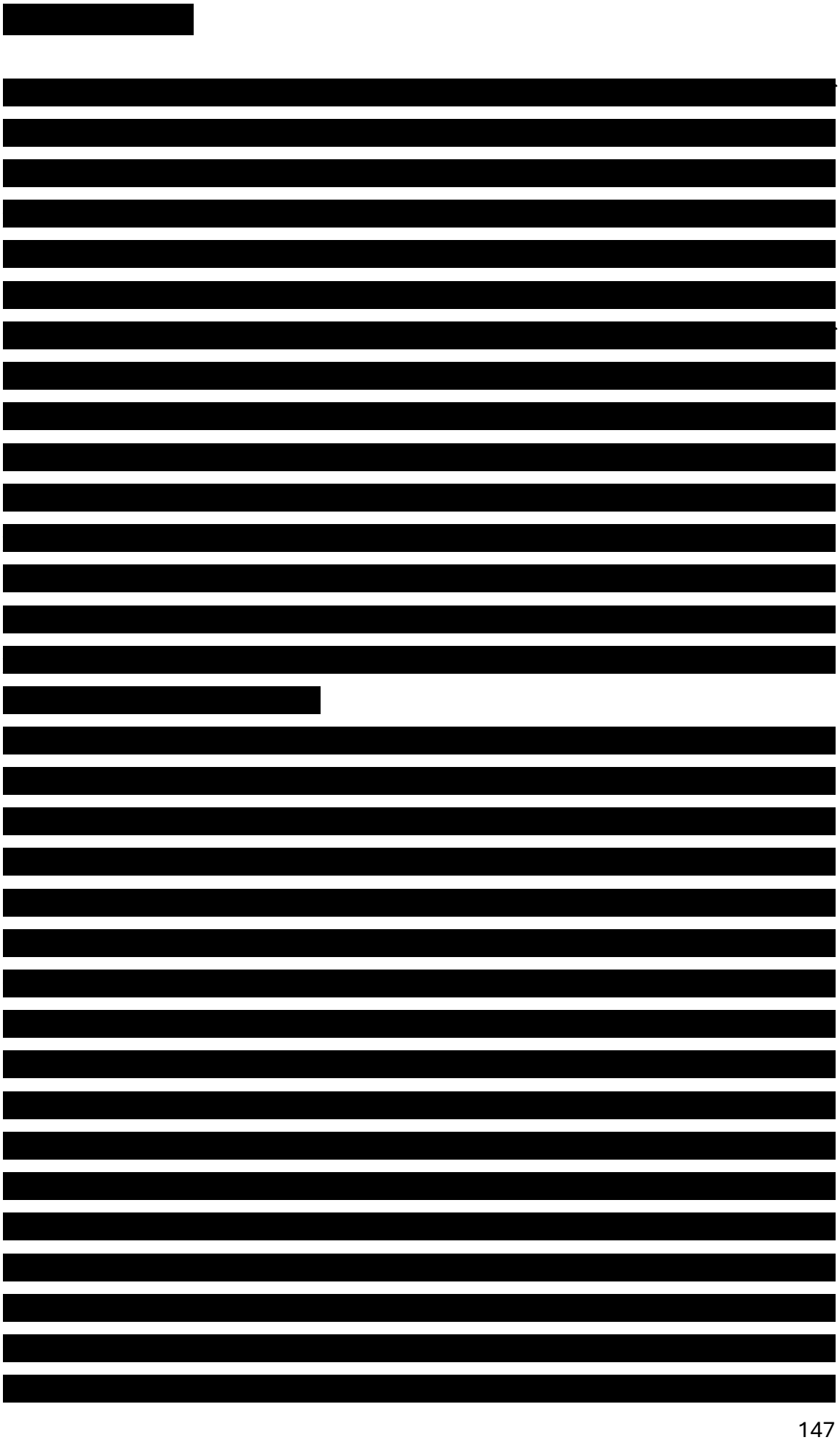

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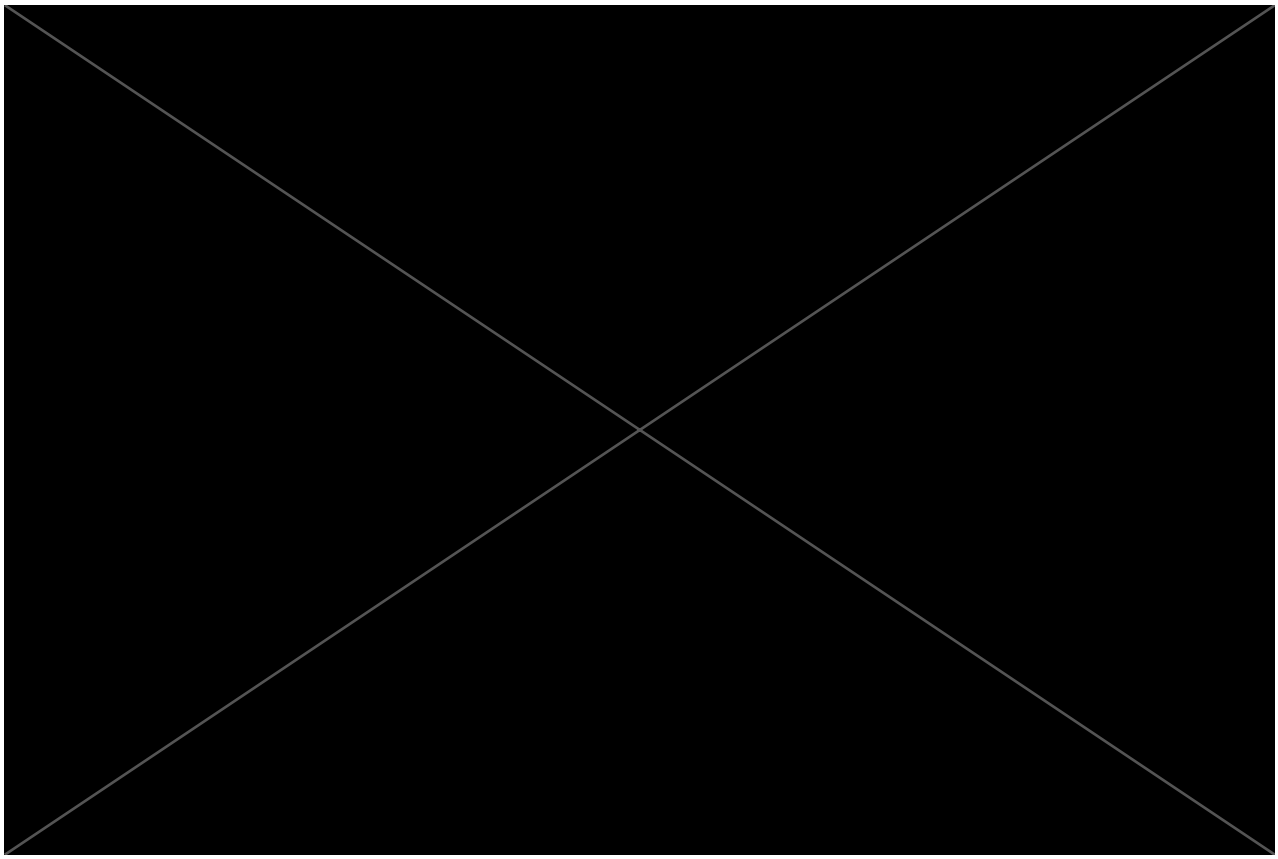

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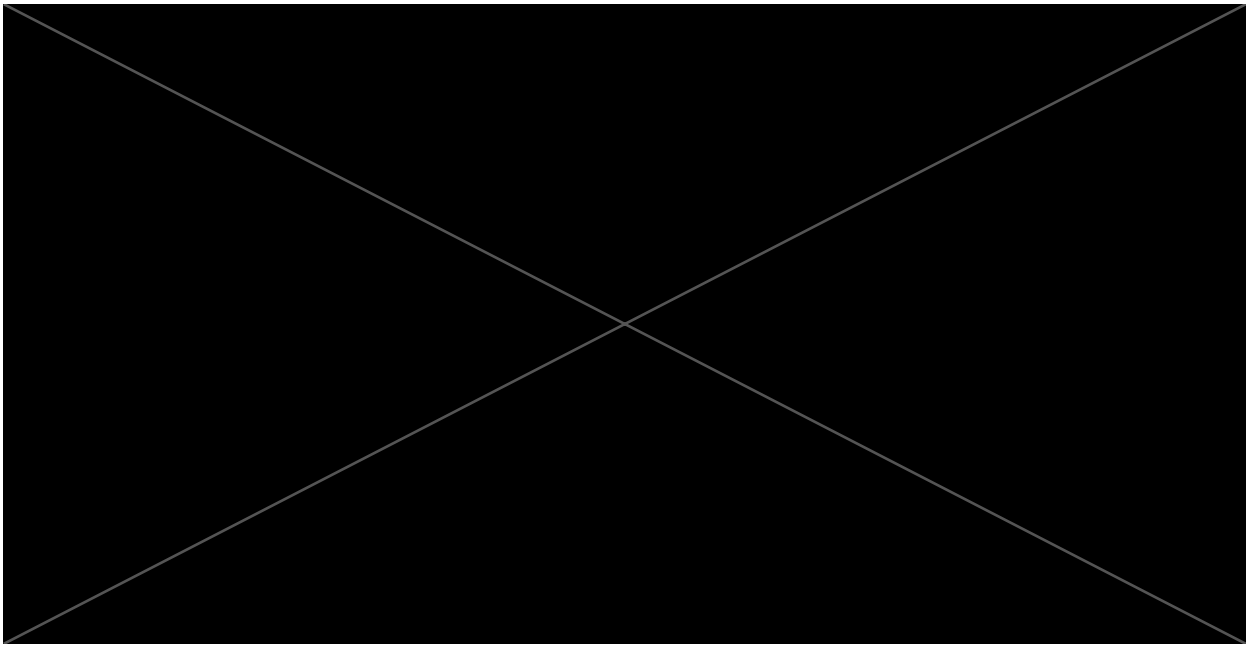

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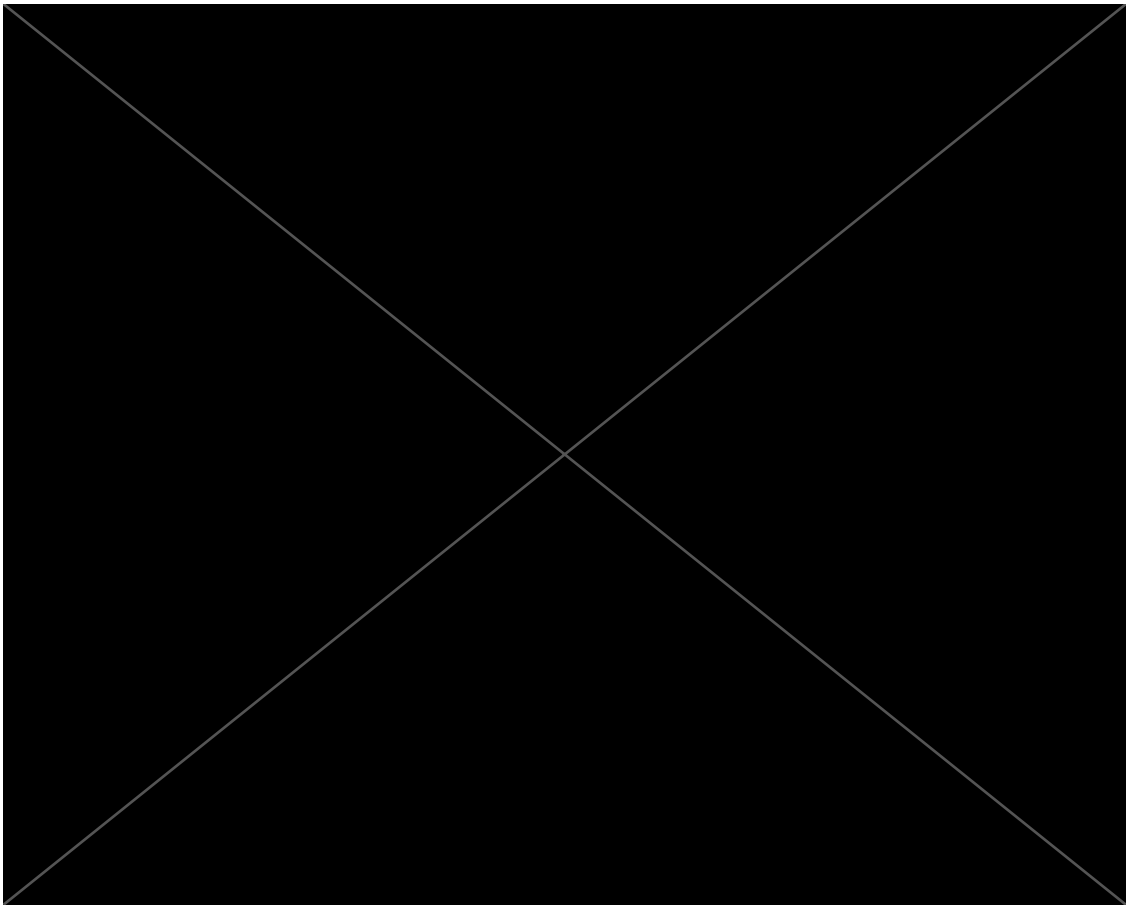

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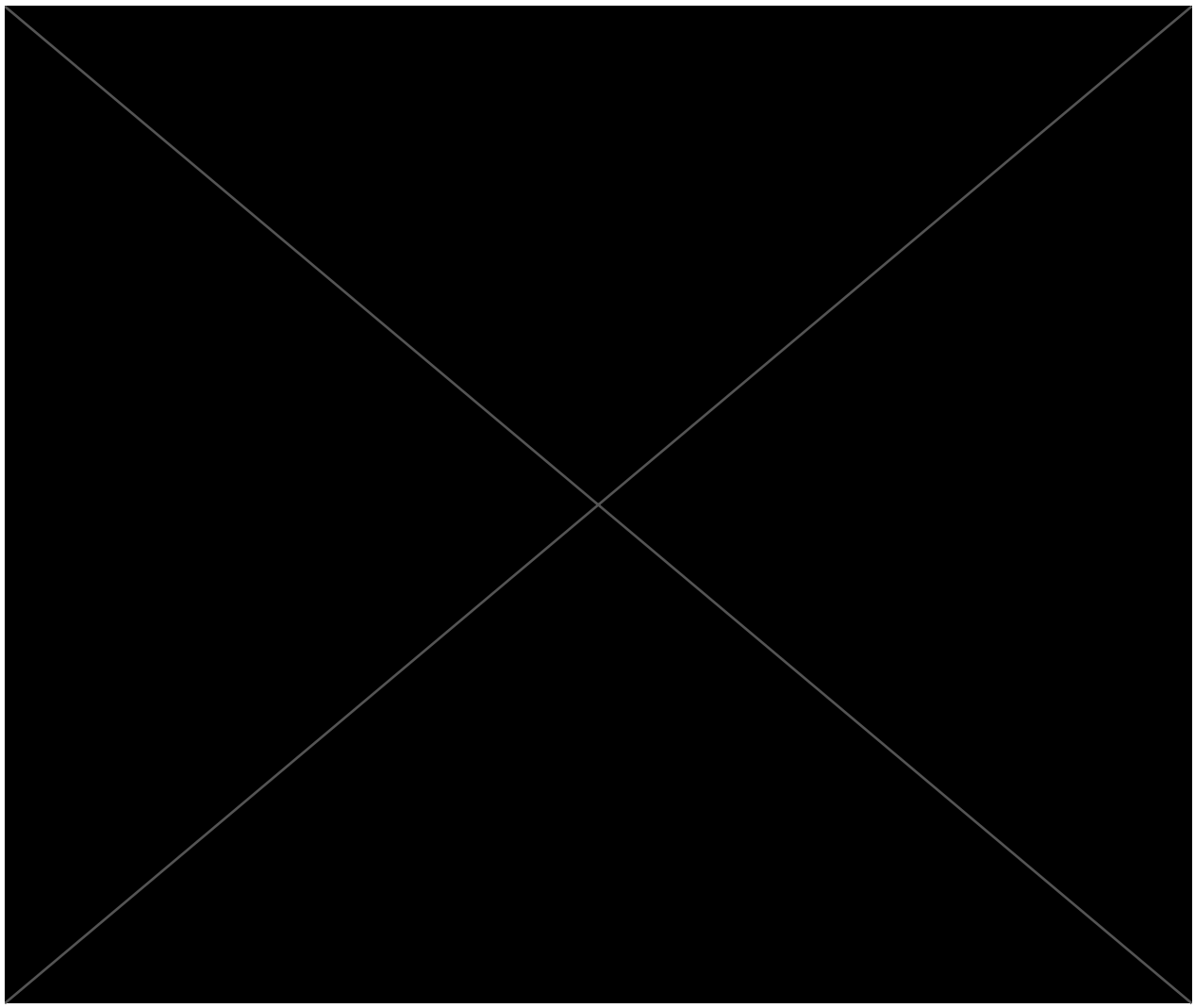

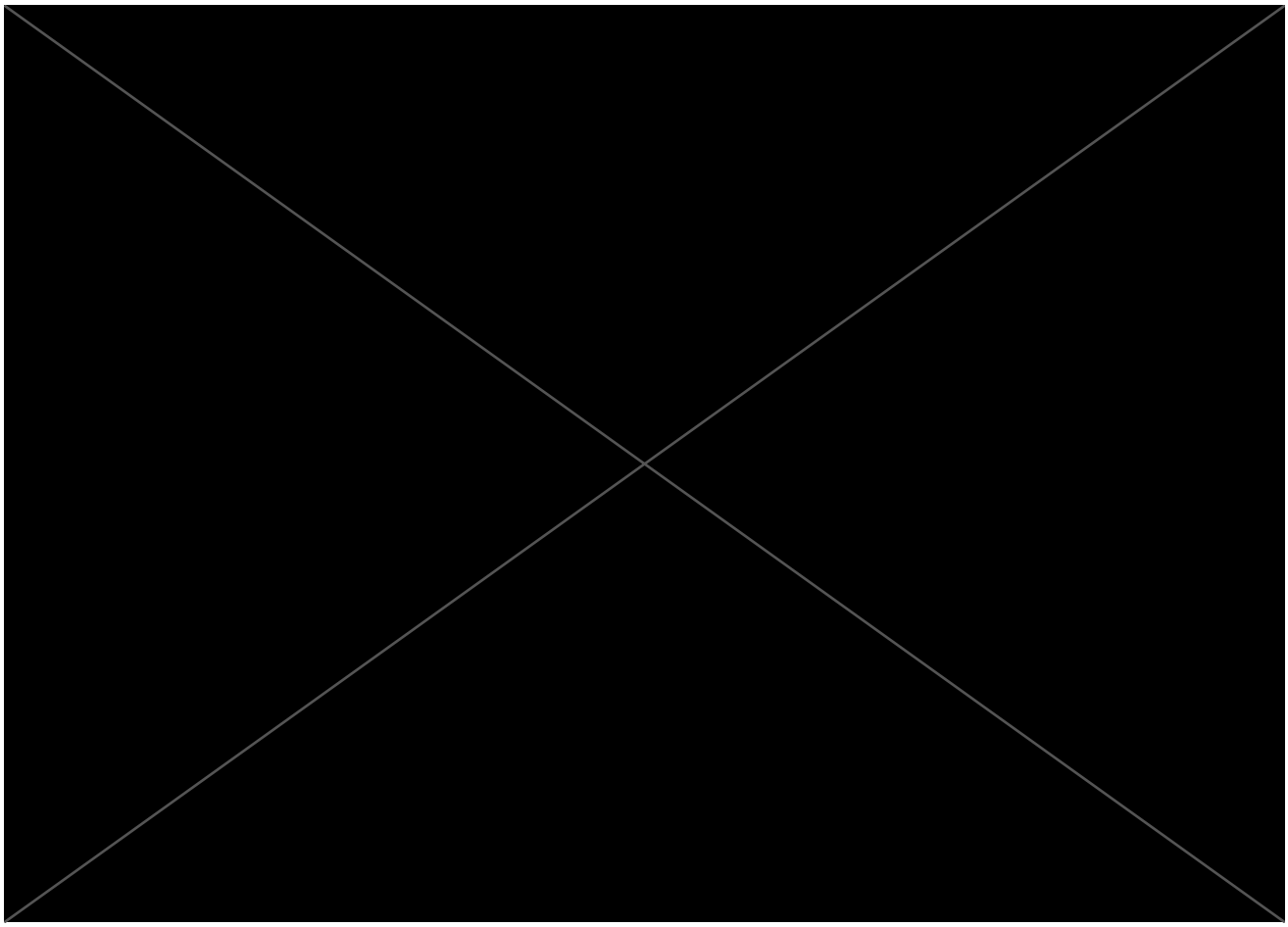

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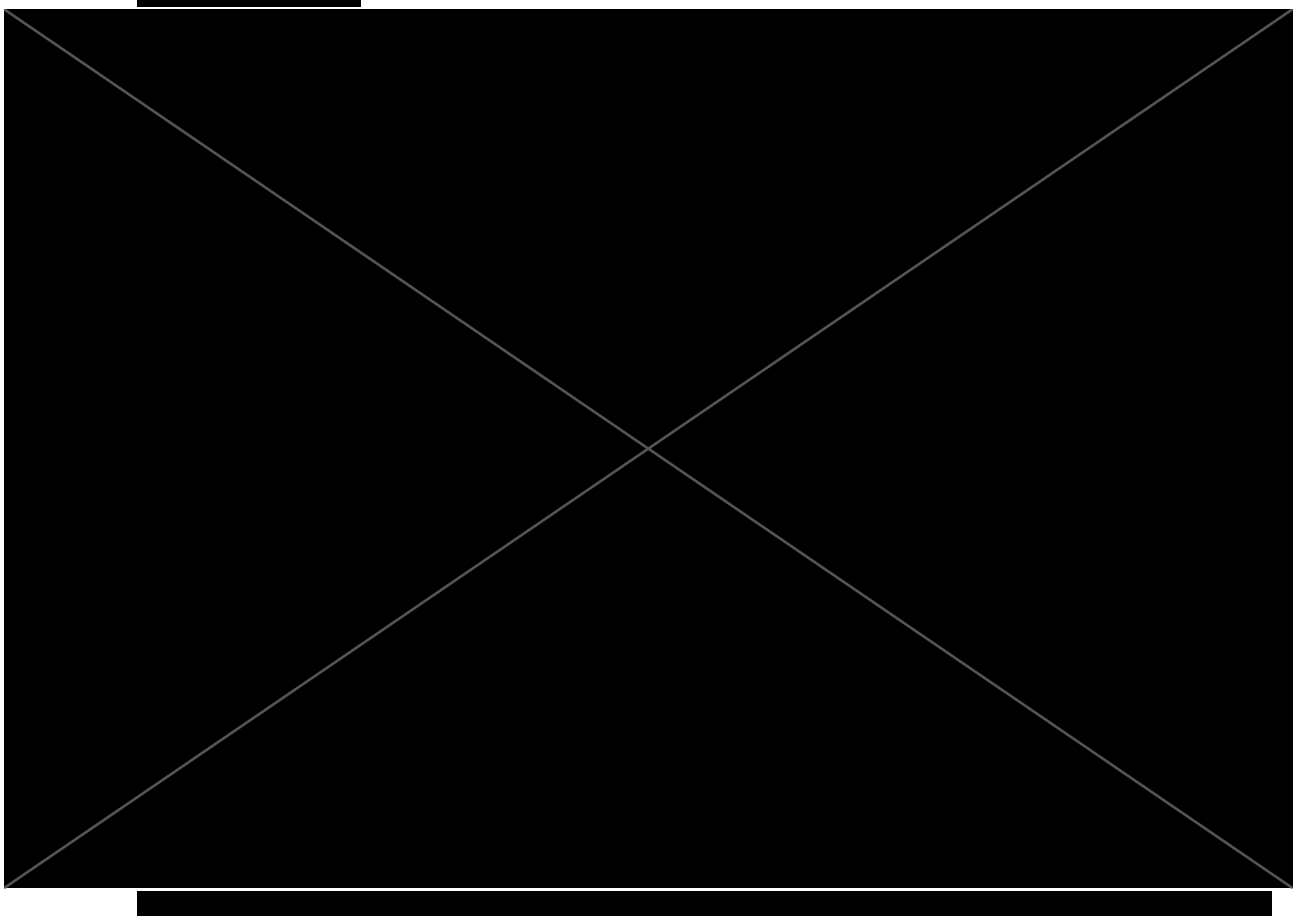

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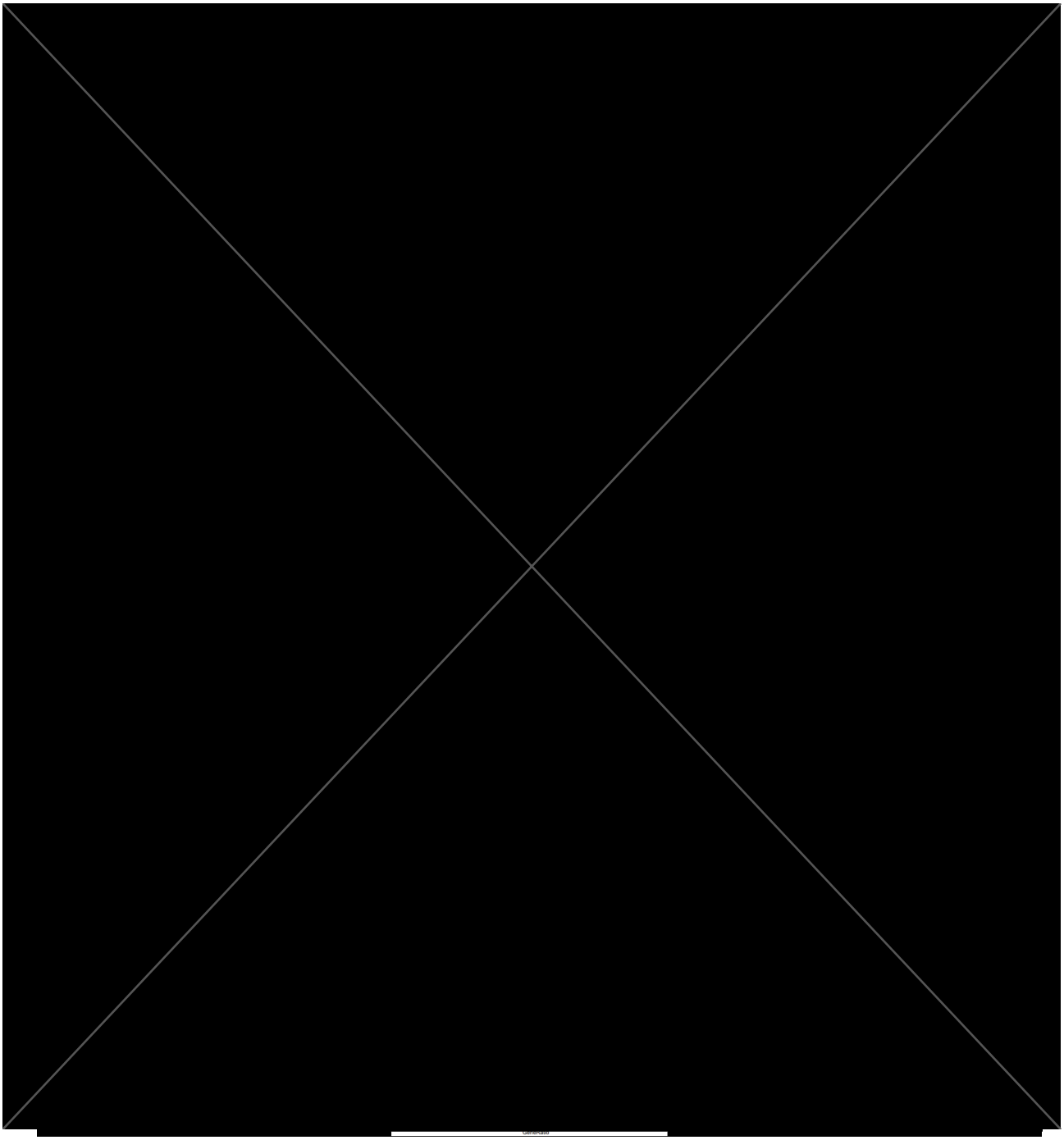

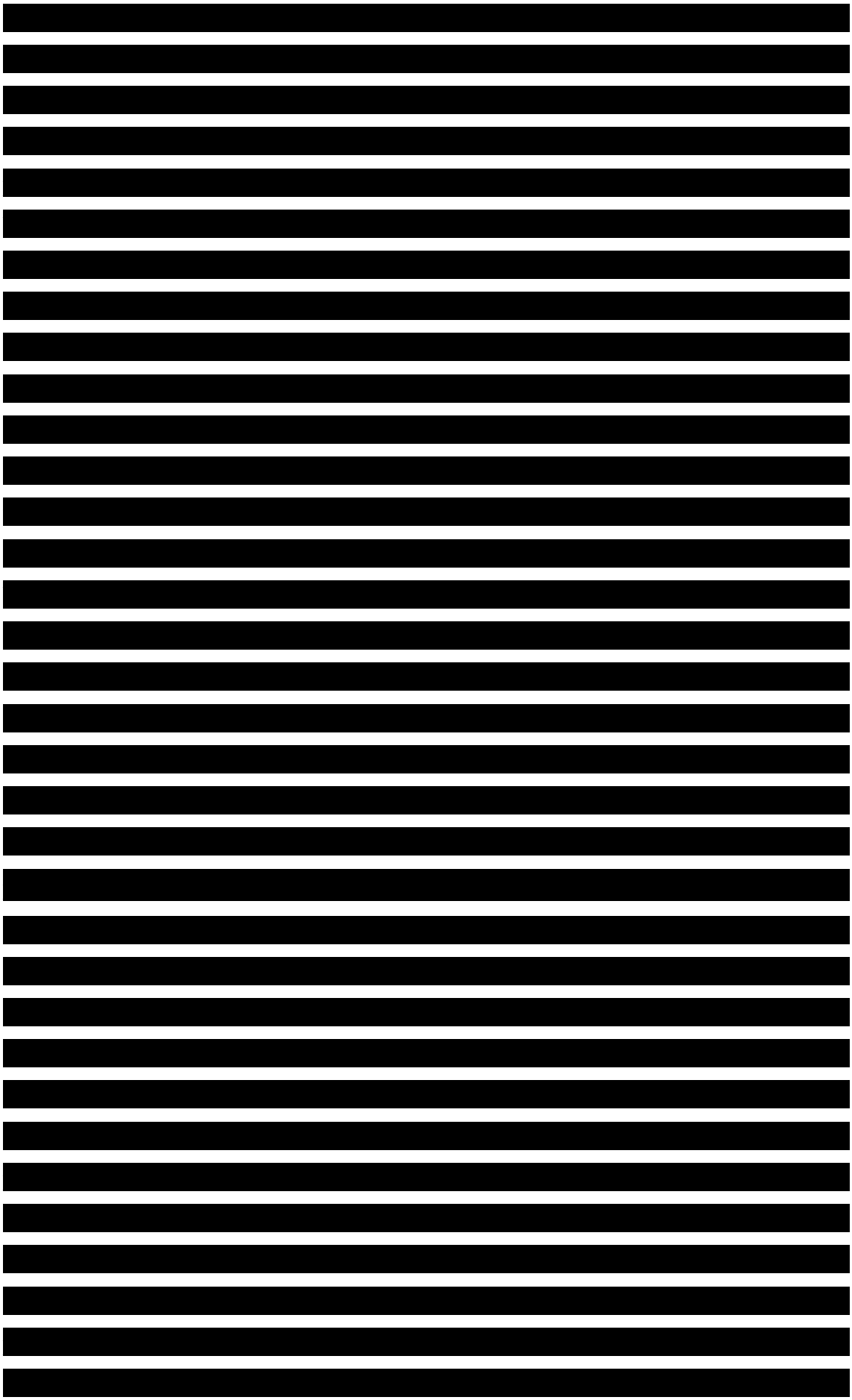

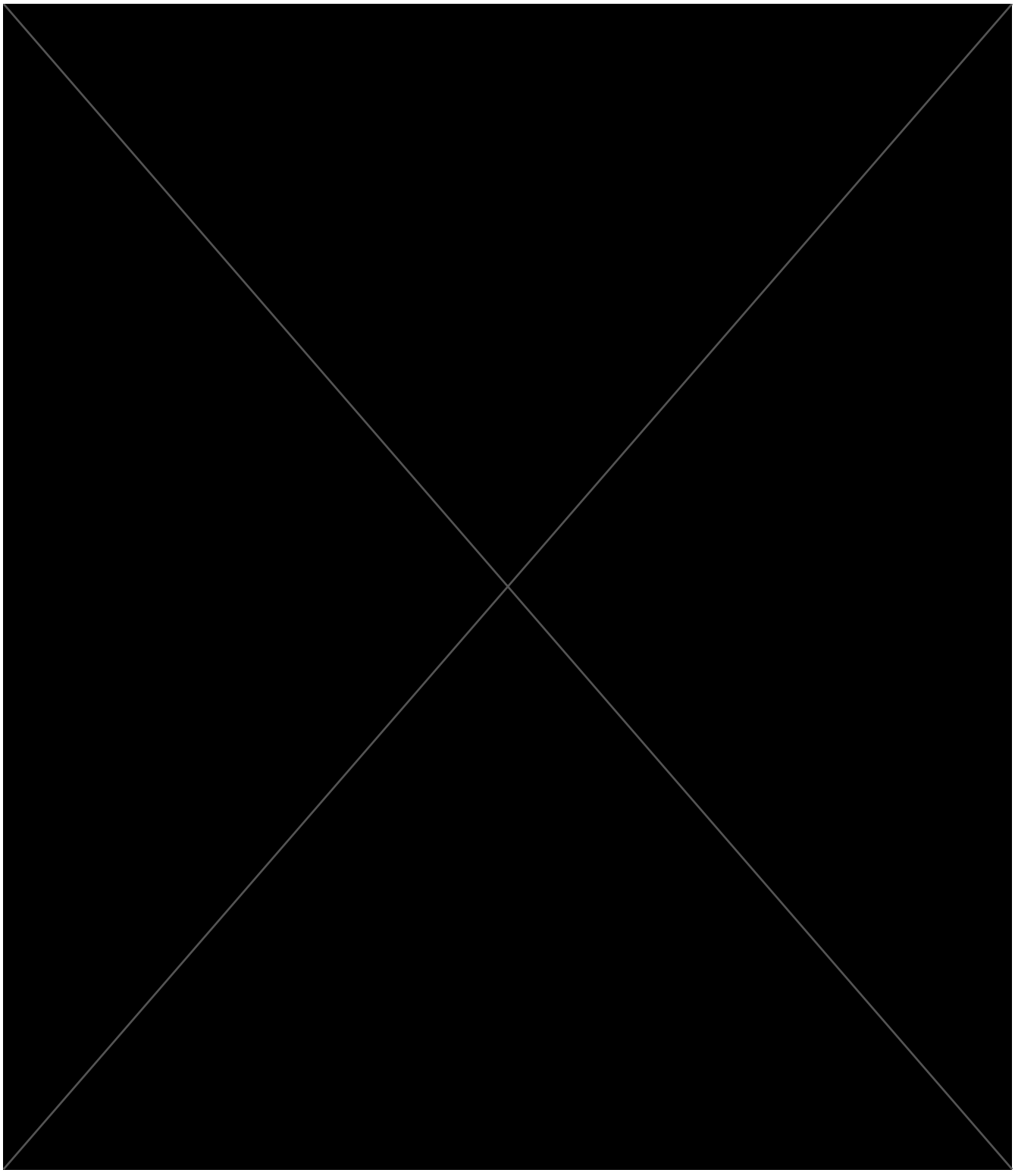

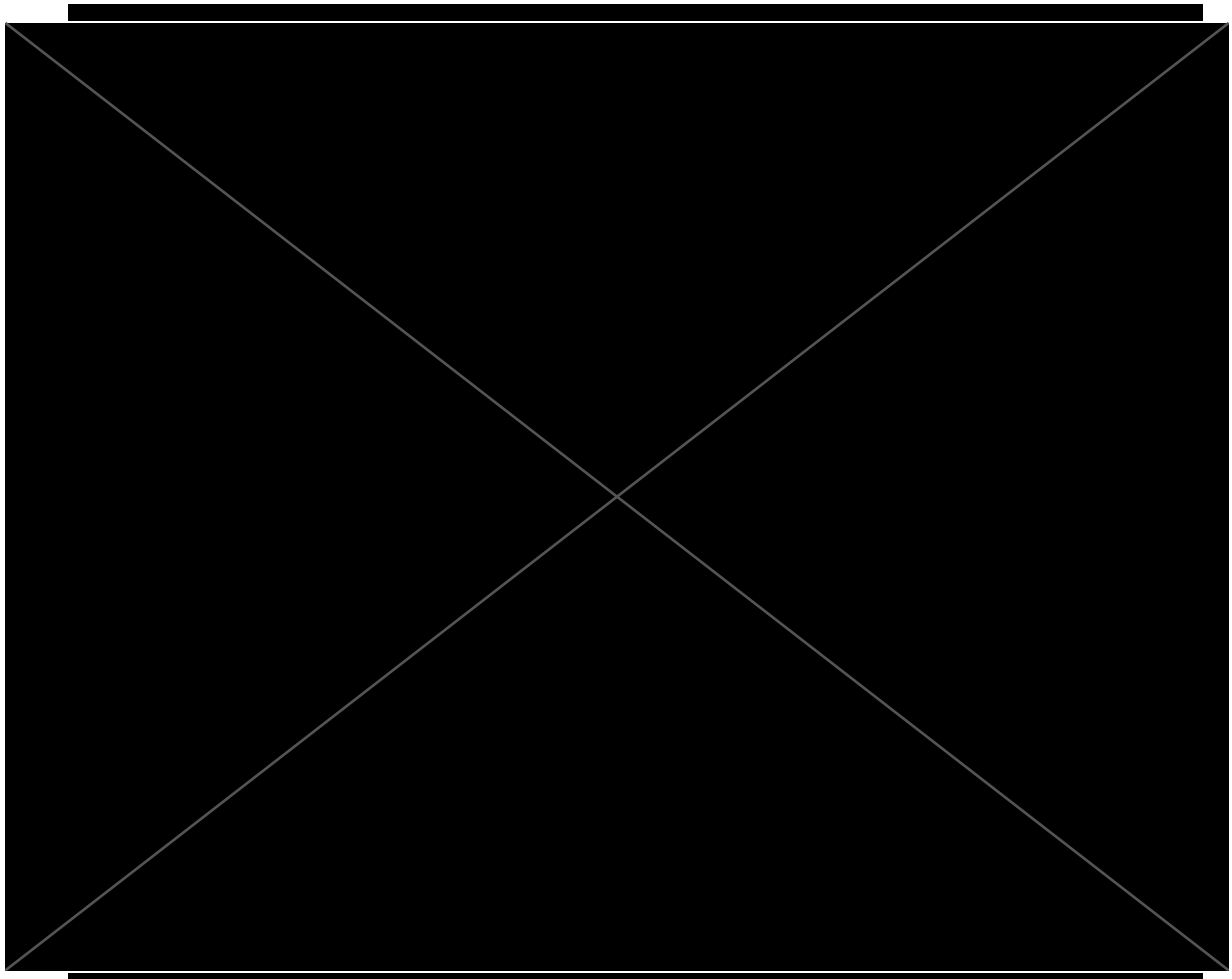

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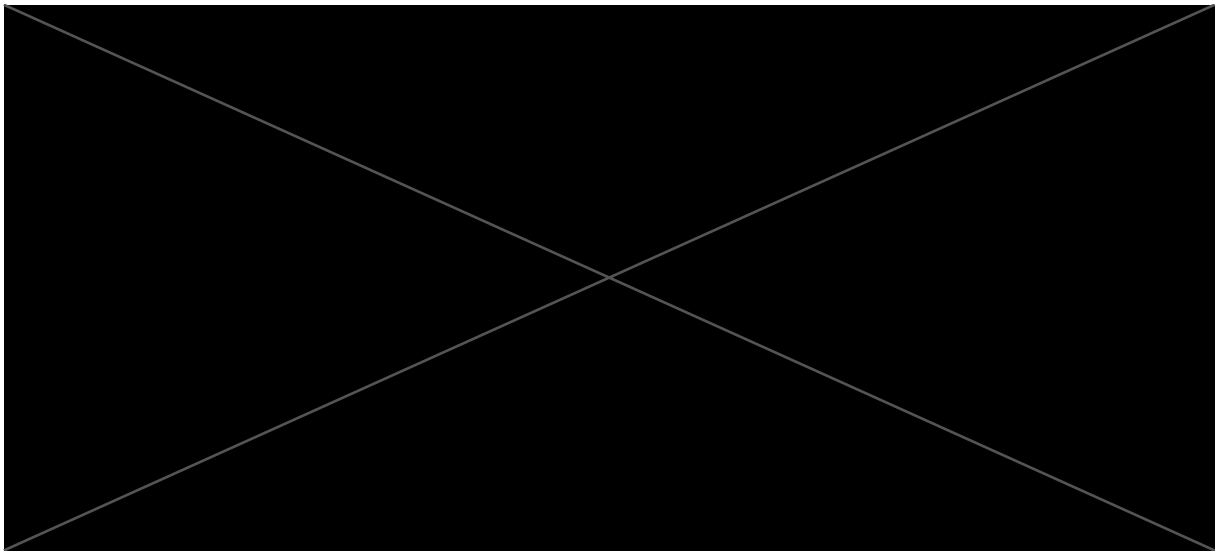

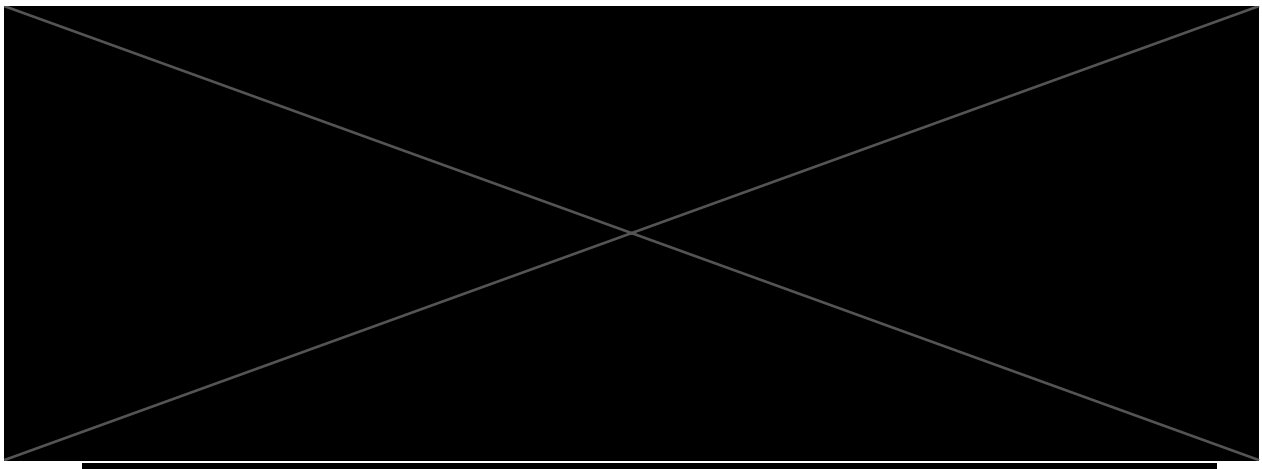

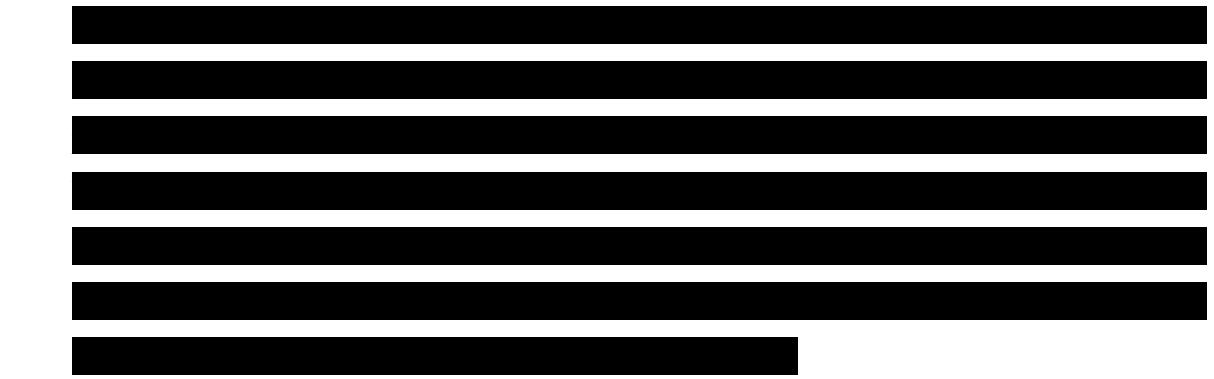

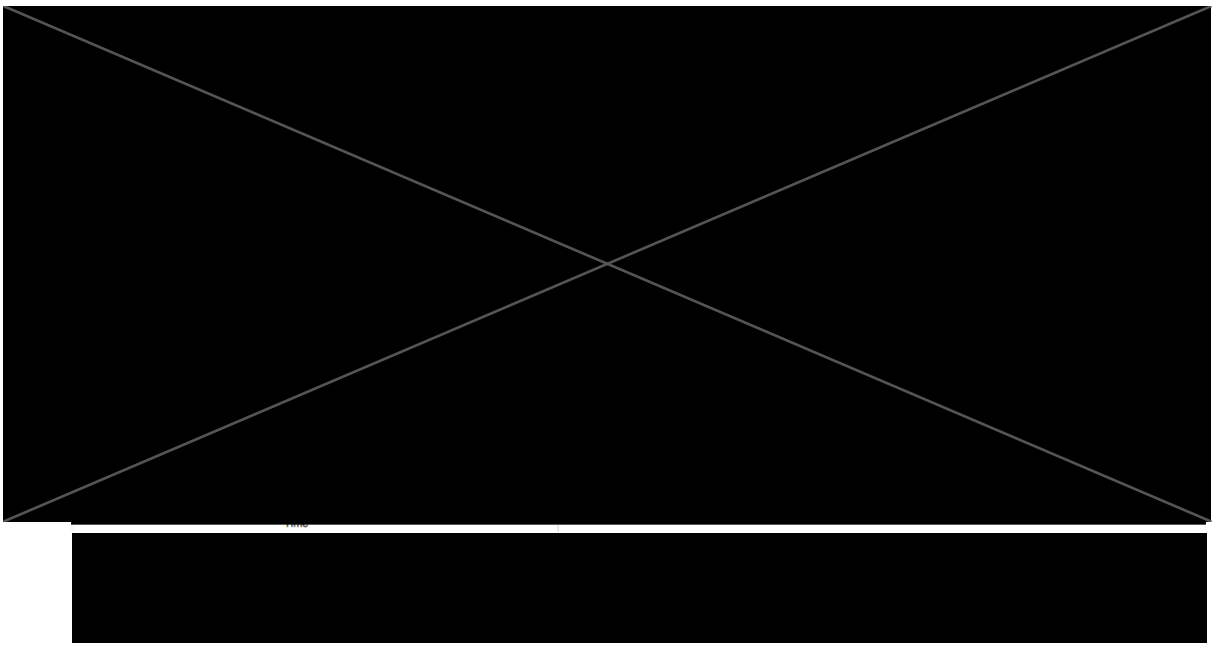

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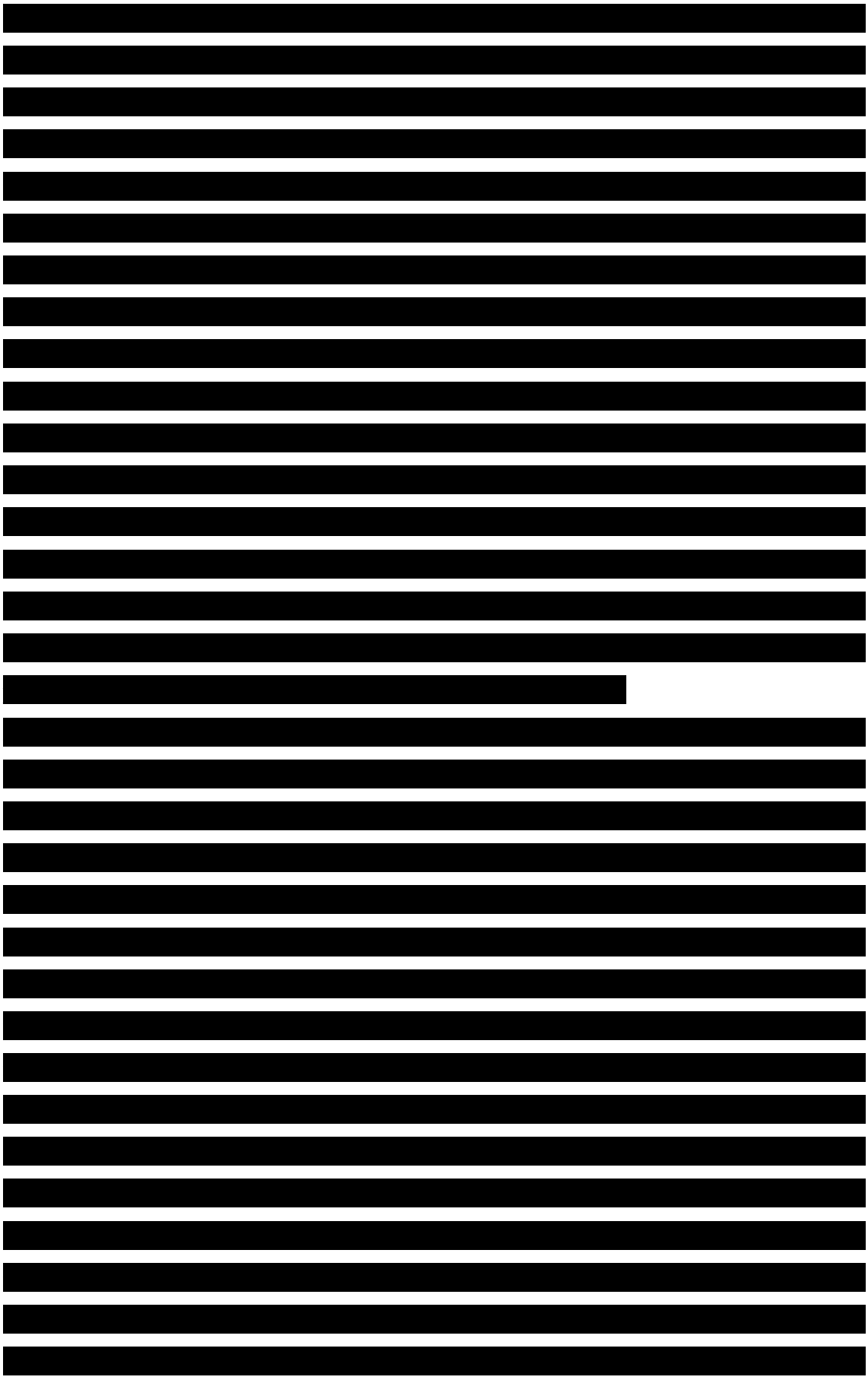

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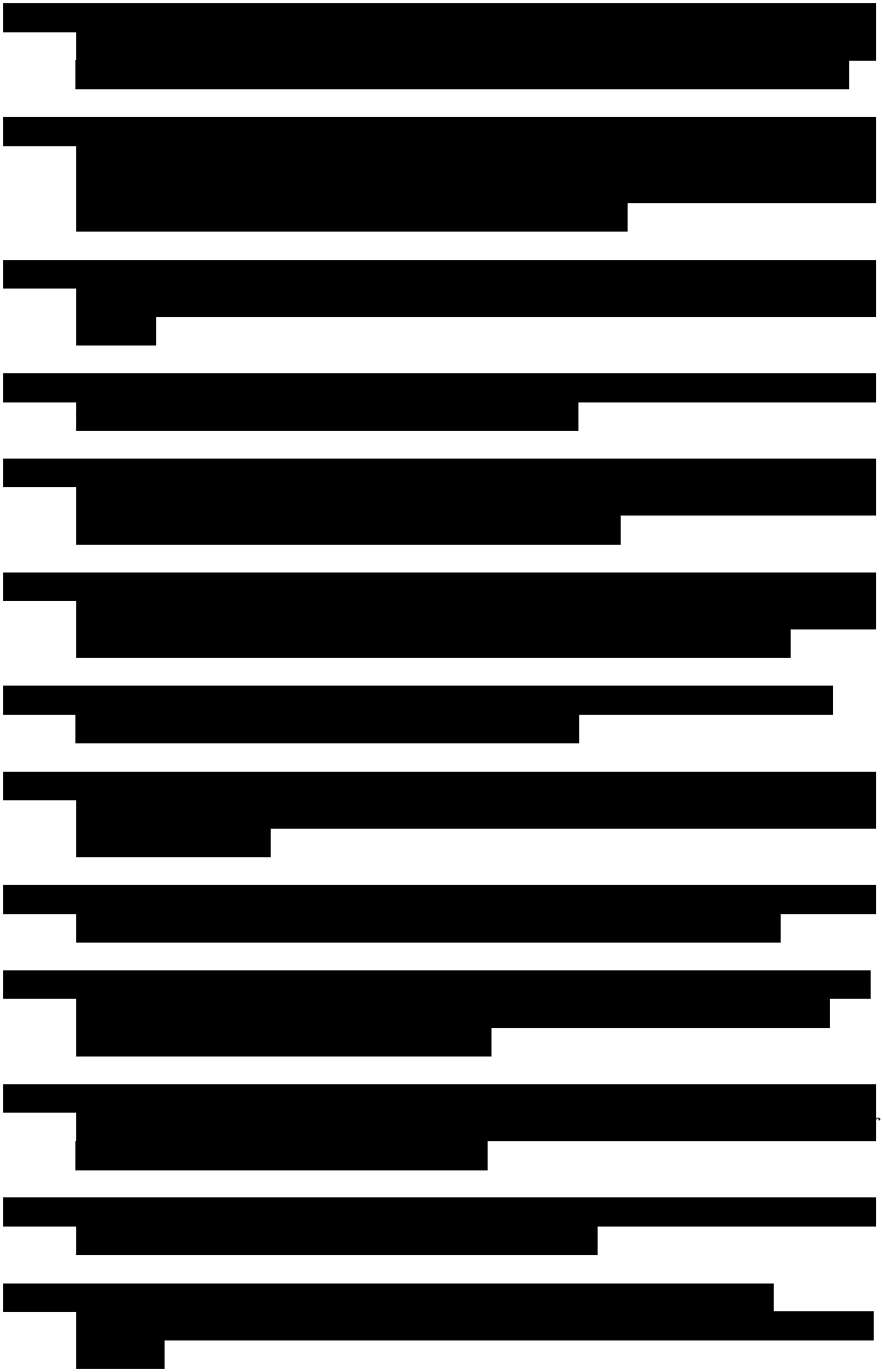

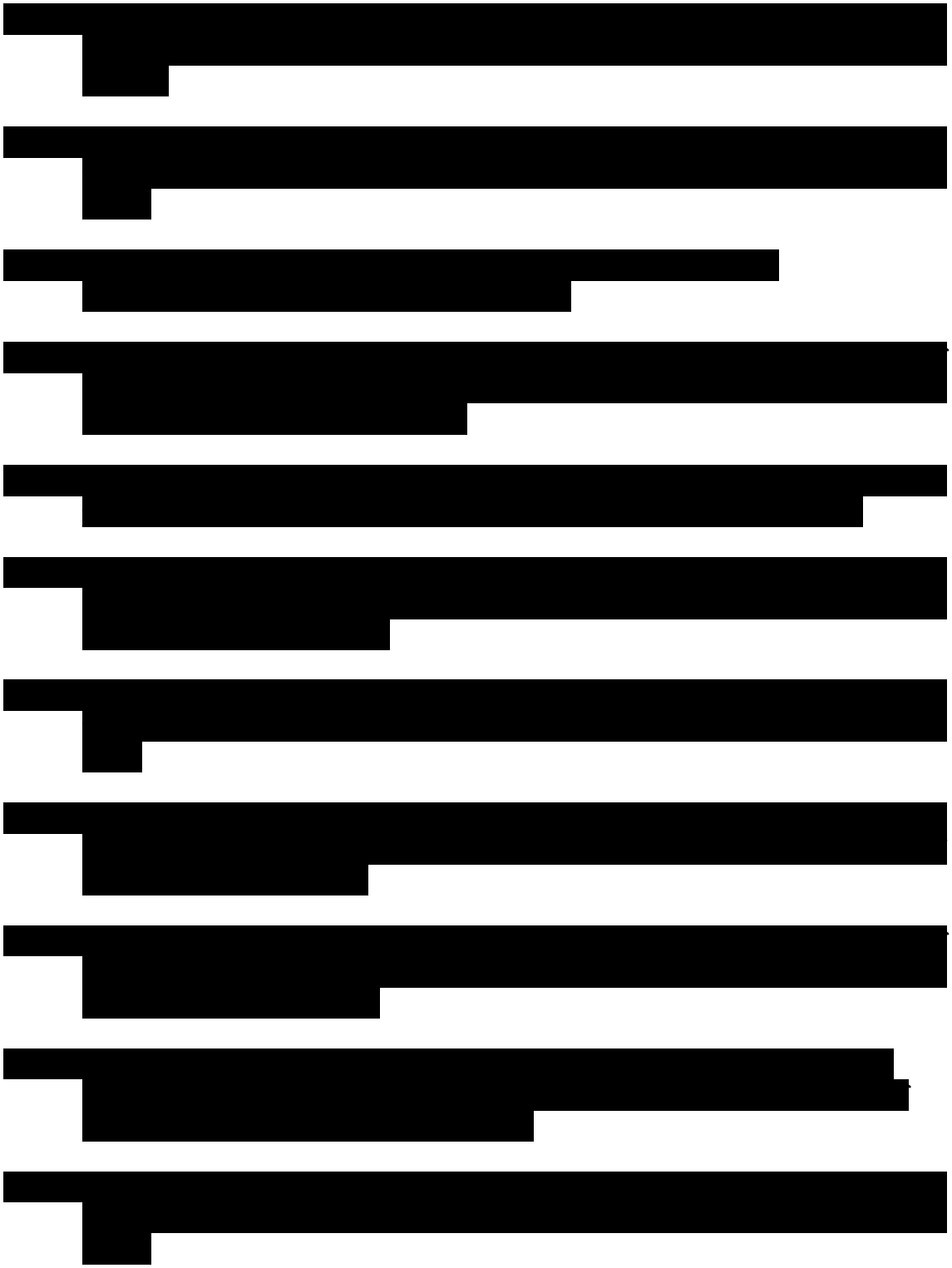

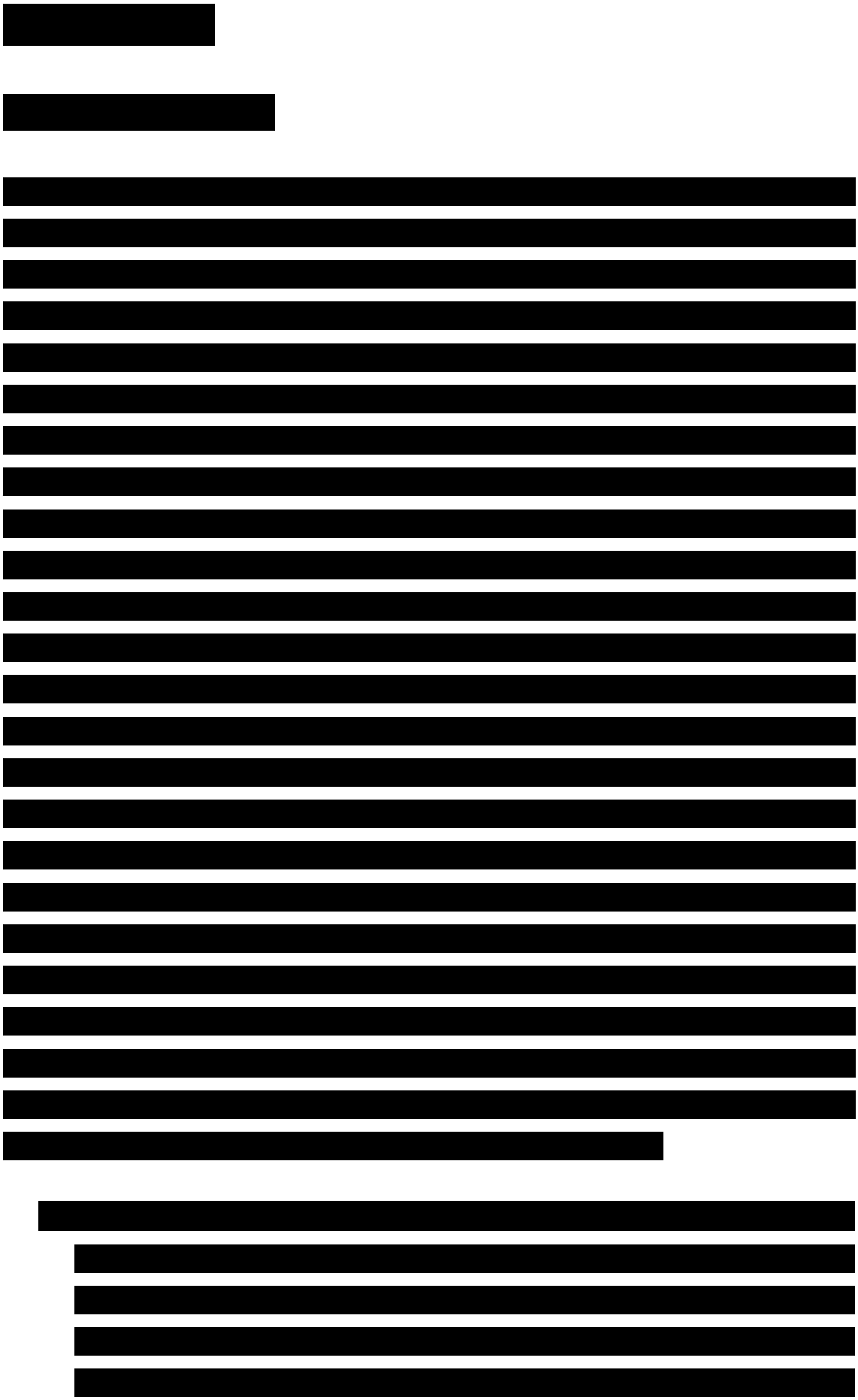

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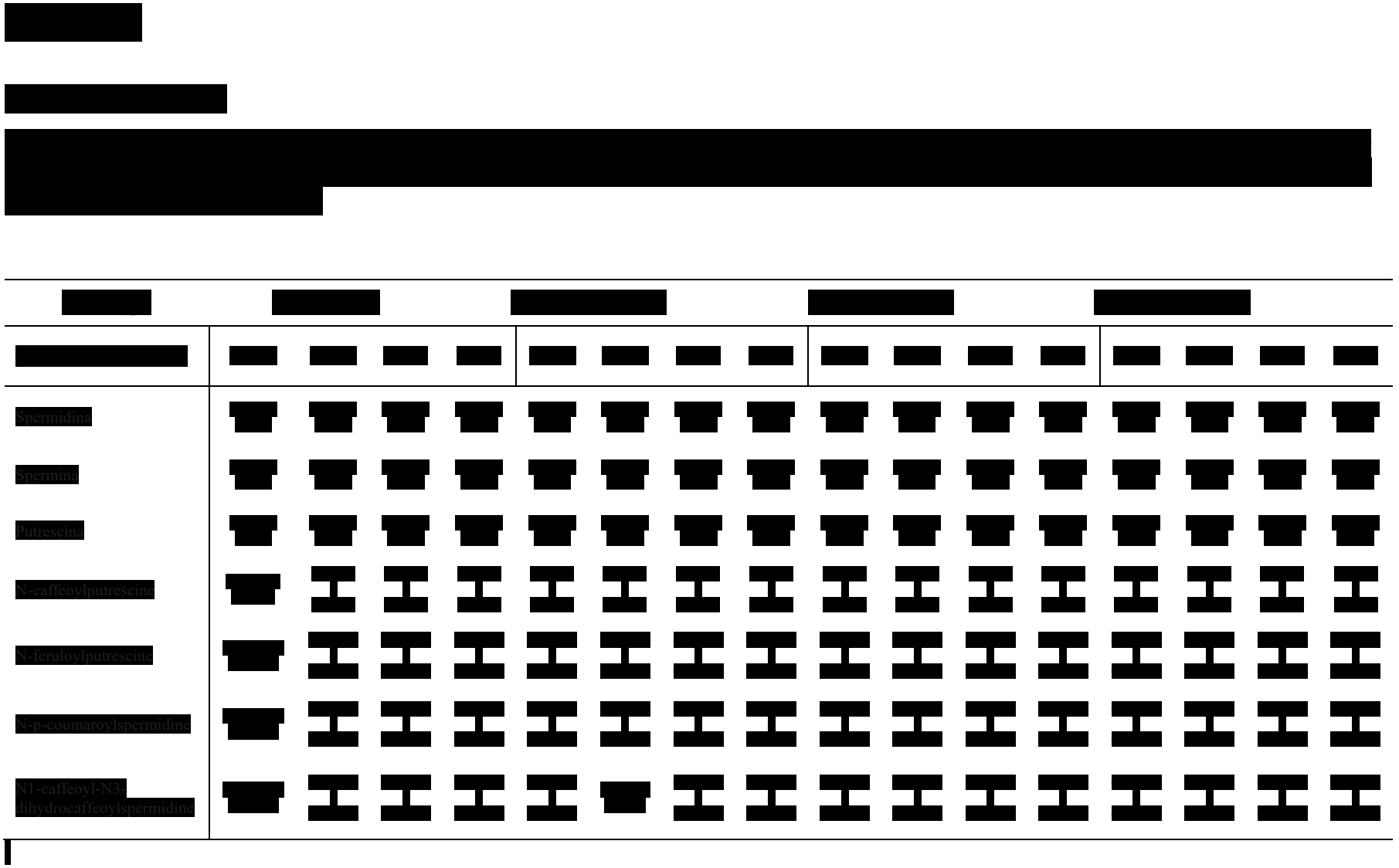
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Spermine
Putrescine
N-caffeoylputrescine
N-feruloylputrescine
N-p-coumaroylspermidine
N1-caffeoyl-N3-
dihydrocaffeoylspermidine