HETEROCHROMATIN, SOMATIC "CROSSING-OVER" AND THE INTERCHANGE HYPOTHESIS BETWEEN NON-HOMOLOGOUS CHROMOSOMES.

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The development of genetics and especially of cytogenetics during the last three decades proceeded so rapidly and the accumulation of differential data in carrying out analytic investigations in these fields of biology was so enormous that it is almost impossible or, better to say, it is very difficult even for a geneticist to keep in contact with all of the trends of the numerous subdivisions in genetics. It is to regret that only a few geneticists work successfully with Drosophila and plant objects, while for the great majority of plant geneticists Drosophila symbolizations are enigmas at the same time when most of Drosophila workers judge the genetic and cytogenetic works carried out with various plants as attempts "to ape the manners of the leaders". The analytic studies in genetics and cytogenetics develop so rapidly that the enormous accumulation of numerous facts in various subdivisions cannot be easily "assimilated" and synthesized. Synthesis is lacking especially between the data accumulated in plant cytogenetics with those of Drosophila studies.

It is true that lack of integrity in genetics as in other biological sciences is often the cause for spending an enormous amount of time and energy on experimenting in certain directions without attaining a satisfactory reward. When integrating, new hypotheses and theories emerge which can further stimulate research work.

Presenting here the data of a series of the observations made by the author in correlation with the numerous observations recorded during the last 2–3 decades upon: (1) heterochromatic regions in the chromosomes, (2) somatic crossing-over, (3) variegations (including mosaicism), and (4) the cytogenetic effect of X-rays, radium-rays and temperature treatments, attempts will be made to advance working hypotheses which will help to link numerous cytogenetic phenomena appearing at first sight as they would have nothing in common. A large number of data, most of which are considered here, suggest that certain environmental factors (X-ray, radium,
temperature, etc.) and internal conditions (conditions in the salivary glands, species hybridization, etc.) apparently induce various degrees of chromosome conjugation, and chiasma formation (crossing-over, interchange) sometimes preferentially between the heterochromatic regions of homologous and non-homologous chromosomes. These phenomena are probably responsible in the majority of the cases for the chromosomal interchanges resulting in the experiments and in nature. Chromosome "fragmentations" are results of such interchanges. Direct fragmentations are probably very rare. Variegations (including mosaicism and some types of chimeras) result in the majority of the cases most probably from somatic interchange following conjugation preferentially in heterochromatic regions from homologous or non-homologous chromosomes. Increased somatic "mutation" frequency in hybrids in many cases is an increase of such interchanges. Agents that induce cancer—most probably induce first chromosome conjugation preferentially in the heterochromatic regions and interchanges in soma between homologous and obviously between non-homologous chromosomes. The same conditions seem to exist in certain genotypes. The observations reported by numerous investigators are given down for supporting these main theoretical interpretations and some others advanced in this paper.

The terms heteropyknose, heterochromatin and chromocenter are known for a long time in the cytological literature, but their possible genetic significance was only recently revealed by the pioneer work of Heitz (1928-35) and Drosophila cytologists (Painter and Stone, 1935; Frolova, 1936, Prokofieva, 1935, 1937; Kaufmann, 1937, etc.).

The cytologists of the last century and the beginning of the present century have often observed in the nuclei numerous bodies which gave usually microchemical reactions like the basic chromatin. Flemming (1882) called them "Netzknoten", Auerbach (1890)—"Cyanophile Nucleolen", Rosen (1892)—"Pseudonucleolen", Zacharias (1895)—"Nebennucleolen", Zimmermann (1896)—"Chromatinkugeln", Telyesnicki (1904)—"Nucleosomen", Rosenberg—(1904-09)—"Prochromosomen", Baccarini (1908)—"Chromocenters", Lundegårdh (1910)—"Karyosomen", etc. (cf. Tischler, 1934). Rosenberg's data are of importance. He found that the number of the prochromosomes corresponds to the chromosome number of the plant studied. Rosenberg's statements were confirmed by numerous investigators in studying the pro-chromosomes (chromocenters) of many plants (Overton, 1905, 1909; Myake, 1905; Yamanouchi, 1906; Laibach, 1907; Malte, 1908, 1910; Guttenberg, 1909; Stout, 1913; Schussning and
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Olde, 1927; Schiller, 1928; Kuhn, 1929; Geitler, 1928; de Souza Violante, 1929; Heitz, 1929; Grégoire, 1932; Janaki Ammal, 1932; Martens and Vandendries, 1933; etc.)

Rosenberg (1909) also called the attention to the inconstancy of the chromocenters (prochromosomes). The inconstancy of the chromocenters was also noted by Tischler (1910) in studying an East-African Musa sapientium. He found that this plant has about as many chromocenters as the half of the chromosome number. Similar observations were reported by Suessenguth (1921) in Dioscorea sinuata and in Thalia dealbata. Tischler interpreted this phenomenon by assuming a secondary fusion of chromocenters from homologous chromosomes (cf. Tischler, 1934). The studies by Lundegårdh (1913, 1914) on Vicia Faba and Allium, by Sharp (1913) in Vicia Faba, and by de Smet (1914) in Crepis showed that the chromocenters concentrated into one side of the nucleus. The investigations by Vejdovsky (1926–27) and especially those by Heitz (1928 a, b, 1929, 1932 a, b, 1933 a, b, 1935) and Grégoire (1907, 1932) disclosed the causal behaviour of the chromocenters tracing the chromosomes from the previous cell division. Grégoire (1907) suggested first that the chromocenters represent those parts of the chromosomes which are lying near the centromeres. The appearance of the so-called "Kappenchromocentren" (Heitz) suggests the possibility of close association of more than two chromocenters, even from non-homologous chromosomes. Heitz and others showed that in some mosses sex chromosomes stain in the resting nuclei like the chromocenters. He assumed that they are inert, i.e., their chromatin substance is "heterochromatic", while those that do not stain are active (euchromatic). In the somatic metaphase plates of highly distained haematoxyline preparations heterochromatin remains still deeply stained, while the euchromatin distains at first, thus the proximal regions (near centromeres) appear darker and the other parts—lighter. Heterochromatin cannot be distinguished by Feulgen reaction. It stains like the euchromatin. Various agents induced a more striking appearance of the heterochromatin in the resting nuclei. The chromocenters are especially strikingly pronounced in actively functioning cells which tend, so to say, to react, to the foreign inductors. Drosera tentacles represent one of the best examples in this respect, that were thoroughly investigated by Hnie (1896, 1897, 1899) and by Rosenberg (1899, 1909). They found that in "resting stage" prophase-like chromatin figures are formed. Similar data were reported by Nicolosi-Roncati (1912) for Pingui-cula hirtiflora and by Faber (1912) for Nepenthes. Similar phenomena were induced by Němec (1910) treating plant tissues by chemicals and
especially "diakinesis"-like nuclei were found by Winge (1927) in crown galls on *Beta vulgaris* which show clearly that abnormal conditions (in this particular case—the conditions created by the infection of beet tissues by *Bacterium tunefaciens*) may induce somatic "conjugations" and, I shall add, perhaps "crossing-over" in soma.

Significantly pronounced chromocenters have been observed in the cells of somatic tissues infected by *Mikorrhiza fungi* (Magnus, 1900; Shibata, 1902; Némec, 1910) and by *nodule bacteria* (Paratore, 1899, 1901; Wendel, 1917; Dangeard, 1926; Kostoff and Kendall, 1929; Kostoff, 1930; Hocquette, 1930, etc.) in *Papilionaceae*. Faber (1912) found that in the cells of infected leaves of *Paveta* and *Psychotria* by bacteria "dicke chromosomenähnliche Gebilde auftreten, die sich stark mit Anilinfarbstoffen tingieren. Diese hyperchromatischen Kerne erinnern so an diejenigen die den Prophasen der heterotypen Teilung zu beobachten sind".

In plant galls, induced by various animal parasites, one can find, in the proximity of the parasites, cells with swollen nuclei having very pronounced chromocenters. I shall recall here the publications by Molliard (1897, 1900), Tischler (1901), Kostoff and Kendall (1929, 1930), Kostoff (1930 and, unpublished), Kendall (1930), etc. In these cases multinucleation is also accompanied with chromosome doubling and an increase of the number of chromocentres. Similar phenomena have been observed in the callus of heteroplastic grafts by Kostoff (1929-30) and in the endosperm produced following interspecific hybridization (Kostoff, 1930, unpublished). Jamaha (1927) observed such phenomena in treating *Vicia Faba* by various chemical agents and Kendall in treating *Pisum sativum*. Yamaha (1927) induced the same cell reaction in exposing *Vicia Faba* at 38° C. temperature. Somatic pairing of chromosomes and somatic chiasma formation were induced by Peto (1935) in *Hordeum* by high temperature. Chiasma formation in somatic cells was induced by Hearne (1936) in treating animal cells with cancerogenic agent methylcholanthrene.

Special attention should be called to the nuclear and the chromosome reactions to X-ray treatments. Pekarek (1927) found that "das Erscheinen von chromocentrischen Gebilden...als wesentlichste Veränderung hervorgehoben ist", in X-raying *Vicia Faba*. Mavor and Svenson (1923, 1924) and Muller (1925) found that X-ray treatments increase the frequency of crossing-over in *Drosophila* near the centromere (spindle fibre attachment) and no change or even a decrease in the more distal parts. Cytological investigations by Heitz (1933, 1935), Painter and Stone (1935) Prokofieva (1936, 1937), Frolova (1935, 1936), etc., indicated that heterochromatic
segments are present at both sides of the centromeres. Correlating these cytological studies with earlier genetic and cytogenetic studies (Dobzhansky, 1929, 1930, 1932; Painter, 1931; Muller, 1932; Muller and Painter, 1932), upon the lack of fitness of the genetic chromosome map in Drosophila with the cytological one, which indicate, that the chromosomes have "inert" regions, i.e., without genes, or with inert, inactive genes (cf. Muller, 1932; Heitz, 1935) some cytogeneticists called the heterochromatic regions as "inert" regions. (Some geneticists as Schultz, 1936, for example, called the term "inert"—a misnomer, considering the fact that inert chromosomes or chromosome segments have occasionally single genes, nevertheless this term is recently more often used in the cytogenic literature than "heterochromatic").

Heterochromatic regions stain darker than the other regions probably because in the heterochromatic regions the chromatides that stain deeply dark are more closely situated and occasionally—somewhat larger. In studying recently the chromatides in the somatic chromosomes of Triticum monococcum and especially of Tr. Timopheevi after fixing root tips in—platine chloride and commercial formalin fixation and staining in gentian violet I succeeded to differentiate very clearly the chromatids of the somatic chromosomes in chromatides during the early metaphase (Fig. 1). There are some regions in the chromatids with much closely situated chromatides while others have chromatides situated at a much larger distance. Large chromatides get further decomposed into smaller but closely situated. Hence one might conclude that heterochromatic regions stain deeply because the chromatides in these regions are closely situated. In other words, the euchromatic regions have less chromatides per length unit of the chromatid and longer interchromatic sections. The sections between each two chromatides that do not stain with the gentian violet after platine chlorid—formol fixation will be called genoneme. This term was used by Koltzoff (1934) as a synonym for chromonema. Since the latter term is used broadly by cytologists and cytogeneticists and the term genoneme fits quite well to the easily distaining treads between the chromatides, I think, it would be better to apply it for these sections. Koltzoff (1934) wrote: "It is difficult to decide what structure corresponds to the gene—the chromatide or the piece of genoneme between two chromatides. The latter assumption seems to me more probable" (p. 313). The chromatide distribution in the heterochromatic (inert) regions in respect to the euchromatic ones gives sufficient background to advance the theory that the genes are located between the chromatides (Kostoff, 1938).
It would be interesting to discuss further the behaviour of the heterochromatic regions. It should be mentioned first of all, that γ-rays of radium induce similar effects upon the chromosomes and upon their behaviour as Röntgen-rays.

In connection with the above data I shall recall here the experiments by Glass (1932) and Oliver (1932) which showed that the regions of the autosomes (Glass) nearer the spindle fiber and the right end of the X-chromosome (Oliver) of Drosophila are more likely to be "fragmented" by irradiation. These regions are the heterochromatic ones ("inert"). In the salivary glands the heterochromatic regions, i.e., the chromocenters conjugate and form a "common chromocenter" (Painter and Stone, 1935; Prokofieva, 1935, 1937; Frolova, 1935, 1936, 1937, etc.).

Finally, the observations by numerous authors should be recalled which showed that X-ray treatment induce chiasma formation (bivalent chromosomes) in soma in the way high temperature does induce. I shall mention here the work by White (1935) who induced "bivalency" (Fig. 9 in White's paper) in Locusla by X-ray treatment during the mitosis which led to interchanges of chromosome segments that conditioned "fragmentations". Similar cytological figures and chromosome behaviour were attained by Riley (1936, Fig. II, 12) in X-raying Tradescantia and by Levan (1937) in X-raying Allium.

All the above given statements and a large number of the same kind, a part of which will be considered later, can be generalized in the following way: (a) The chromosomes of plants and animals have heterochromatic regions which stain deeply during the mitosis and even during the resting stage or prophase (chromocenters). (b) The chromatids during the early somatic metaphase, when properly fixed and stained can be differentiated into chromomeres the latter being much closely situated in the heterochromatic regions. (Regions with chromomere condensations have been found during the early prophase, which probably correspond to the heterochromatic regions.) (c) The heterochromatin is chiefly concentrated near the centromeres but some of the chromosomes have heterochromatic segments on the distal ends too as the observations in Drosophila (Prokofieva, 1935, 1937), Crepis (the author, 1938), Triticum monococcum (the author, 1938 in press) and other objects showed (Fig. 2). (d) External factors (chemical agents, parasites, X-rays, temperature, etc.) induce pronounced appearance of the chromocenters and a reduction in number, which is probably due to conjugation of the chromocenters. When an increase in number of the chromocenters has been observed, two interpretations can be advanced: (1) striking
appearance of the distal heterochromatic ends and (2) the same agents that have induced a pronounced appearance of the chromocenters, have also conditioned chromosome doubling. The occurrence of these two possibilities at the same time is not excluded. Cancerogenic agents (Hearne, 1936), parasites (Winge, 1927), X-rays (Marquardt, 1937), temperature (Peto, 1935), etc., induce chromosome association (often perhaps between non-homologous chromosomes) in somatic cells.

Somatic chromosome conjugation takes place in active glandular cells without any treatment (i.e., in the salivary glands) where euchromatic homologous regions conjugate normally as they do during the meiosis, while the heterochromatic proximal regions of all chromosomes conjugate all together (non-homologous). The investigations by Prokofieva (1937) upon the structure of the chromocenter showed that the inert (heterochromatic) arm of the IV-chromosome conjugates with the heterochromatic regions of III- and II-chromosomes and the heterochromatic region of the X-chromosome (proximal) conjugates with those of IV- and II-chromosomes, i.e., a conjugation of heterochromatic regions of non-homologous chromosomes. Genetic data obtained by Gershenson (in press) in studying the preferential segregation in triplo-IV flies (Drosophila) having XY + "fragmented" X also suggest a conjugation between non-homologous chromosomes probably in the heterochromatic (inert) regions. Kikkawa (1937) observations on the genetic behaviour of haplo-IV Drosophila ananassae suggested a probable conjugation too between the heterochromatic regions of IV-chromosome and Y-chromosome. Meiotic chromosome conjugations, chiasma-formation and crossing-over, as a rule, result from an attraction between homologous parts carrying equal genes, i.e., between like-parts (Jennings, 1923; Morgan, Bridges and Sturtevant, 1925; Muller, 1928; Dobzhansky, 1930, 1931, 1932; Dobzhansky and Sturtevant, 1931; Creighton and McClintock, 1931; Stern, 1931; Chino and Kikkawa, 1933; etc.). Synapsis that takes place between heterochromatic regions (also called inert regions) of the non-homologous chromosomes can be interpreted by assuming a likeness between these regions, no matter that they are distributed in all chromosomes. If we speak with genetic terms, they represent inactive "duplications" that can be found almost in each chromosome at the proximal ends and occasionally on the distal ones. Y-chromosome in Drosophila melanogaster is in its greater partheterochromatic (inert). It is genetically inert except of a few genes. Synapsis during the meiosis in normal diploids is chiefly regulated by the attraction of homologous active (euchromatic) parts. The hypothesis advanced by Snell (1938) for explaining
haploids especially (this species considering hybrids altogether regions induce regions synapsis act homologous gations. in reality, conclusions appear clear of the reality, the terms "homologous" and "non-homologous" chromosomes appear in a new light. The contents of these conceptions evolve. It is now clear why the inert (heterochromatic) B-chromosome of Zea conjugates with itself by "folding back", or why three B-chromosomes conjugate in forming T-like figures as shown by McClintock (1933) excellent investigations. Randolph (1928) accumulated such B-chromosomes more than 25 in a plant without conditioning marked morphological effect. Avdulov (1937) found heterochromatic (inert) regions in the A-chromosomes (the active ones that have genes), which might also account for some "non-homologous" chromosome attractions and associations.

Biochemical and biophysical processes that proceed during the meiosis act in certain organisms somewhat differently on the chromosomes than those in the salivary glands since during the meiosis they usually induce synopsis between homologous chromosomes and between the heterochromatic regions of the homologous chromosomes, while in the salivary glands they induce close synopsis between homologous active parts and act upon the inert regions of all chromosomes in such a way that the inert regions conjugate altogether forming usually a common "chromocenter".

Association of heterochromatic regions in many haploids and species hybrids with asyndesis during the meiosis does not seem to be a rule, considering the fact that most of the haploids (cf. Kostoff, 1938) and many F₁ species hybrids have asyndesis during the meiosis. On the other hand associations between "non-homologous" parts of A-chromosomes, and especially of B-chromosomes in Zea, association of the chromcenters of bivalents in Vicia (Enin, unpublished) forming sometimes a star-like figure (this occurs rarely), and end-to-end conjugations in Triticum monococcum haploids remind of the conditions in the salivary glands.
I wish to call the attention here to some observations made recently which will be of significance for our further discussions. In the F₁ species hybrids Nicotiana bonariensis (n = 9) × N. Sanderae (n = 9) I have found occasionally during the I and II meiotic anaphases one and rarely two chromatin bridges which undoubtedly result from crossing-over between inverted segments. Bivalents that appeared during the I meiosis of these species hybrids had quite often more than one chiasma per bivalent, which accounts for the formation of chromatids with two centromeres and a fragment without centromere during the II anaphase. The tapetum cells (the layer of somatic cells that envelopes internally the pollen sacs in which the pollen-mother cells develop) represent the cell barrier between the soma and the sac inside of which meiosis proceeds, i.e., the place where processes proceed that induce chromosome pairing (cf. Kostoff, 1930). Tapetum cells have very active metabolic and catabolic processes. They expand enormously, their nuclei often divide once or several times, while the cells divide rarely; therefore they usually have more than one nuclei or polyploid nuclei: tetraploid, octaploid or even of higher polyploid grades. Normally, no chromosome conjugations take place in these cells. But sometimes conjugations occur. It is occasionally followed by interchange of chromosome segments. I have observed it rarely in pure species, but got the impression that it occurs more frequently in species hybrids. It occasionally occurred in the hybrid N. bonariensis × N. Sanderae as a result of which somatic anaphases with chromatin bridges (i.e., chromatids with two centromeres) have been found (Fig. 3). In this case it is difficult to decide whether the chromatids with two centromeres originate following somatic crossing-over between bonariensis and Sanderae chromosome pairs which in meiosis behave as chromosomes having inversions, or interchange has occurred between heterochromatic regions of non-homologous chromosomes located in such places of the chromosomes, that chromatids with two centromeres get formed.
after the interchange. A hybrid *N. Langsdorffii* (*n* = 9) × *N. Sanderae* (*n* = 9), however, which I studied in 1931, offers a more definite answer. This hybrid formed normally 9 bivalents during the meiosis, while in the tapetum cells I have occasionally found anaphases with bicentric chromatids which can be interpreted by assuming interchange between non-homologous chromosomes.

The exceedingly small size of the fragment formed from interchange (I could not find it at all in most of the cases) in the tapetum cells of *bonariensis × Sanderae* can serve as an argument that crossing-over has taken place in the very distal ends of the chromosomes. The size of the fragments originating from crossing-over during the meiosis between inverted chromosome segments were in most of the cases much larger than those in tapetum. This suggests a possibility of interchanges in tapetum cells between extreme distal ends of the participating chromosomes. Some of the chromosomes have heterochromatic segments at the distal ends. Consequently, one cannot exclude the possibility of interchange between heterochromatic distal ends.

Some of the extensive data reported in the excellent paper by Stern (1936) upon the somatic crossing-over suggest strongly that in soma crossing-over occurs following conjugations between heterochromatic (inert) regions.

Stern (1936) wrote "the relative frequencies of somatic cross-overs in different regions of *X*-chromosomes are different from those of germinal cross-overs. Somatic crossing-over is more frequent near the fibre point. The presence of Minute~*n* accentuates this shift" (p. 727). *X*-chromosome has a large inert region near the centromere. It does not seem improbable to suggest that the inert region is involved in these interchanges. This suggestion is also supported by the following statement made by Stern: "The *X*-chromosome duplication—*θ*—frequently undergoes somatic crossing-over with the *X*-chromosome—more frequently in the homologous right than in the homologous left regions. Germinal crossing-over involving *θ* is very rare."

Gene Minute seems to act in a specific way creating conditions that induce chromosome synapsis in soma and crossing-over in the (or in the proximity of the) inert regions. According to Stern, "Somatic autosomal crossing-over takes place in both sexes, though more frequently in females. A peculiar specificity of the Minute effect leads to cross-overs in that arm of the third chromosome in which the Minute itself is located. Most cros-sovers are concentrated near the fibre point region." The fibre point regions
are heterochromatic (inert). The effect of Minute gene is not great, therefore it is local. But genes with such a kind of effect (although somewhat greater) inducing crossing-over in heterochromatic chromosome regions, especially when it might also proceed between heterochromatic segments of non-homologous chromosomes might be decisive for cancer formation in case cancer results from "crossing-over" (interchange) between non-homologous chromosomes, as it seems to be so. (This problem I shall consider later again.)

According to Oliver (1934) "Somatic pairing is never normal, but is characteristic for the attraction of like parts" (cf. Dobzhansky and Sturtevant, 1931; Oliver and Van Atta, 1932; Sturtevant and Dobzhansky, 1930; Van Atta, 1932). The data obtained by McClintock (1933) and Jones (1936) suggest, however, crossing-over between non-homologous chromosomes in maize. In studying somatic segregation in maize Jones (1936) concluded: "The loss of a series of linked genes could be due either to non-disjunction, somatic crossing-over of homologous chromosomes, reciprocal translocations (non-homologous crossing-over), translocation or delation" (p. 166). If we correlate Jones' data with McClintock (1933) observations upon the conjugation of non-homologous chromosomes, which we interpreted above as conjugations between heterochromatin it is quite logical to interpret Jones' data by the assumption of interchange between heterochromatic regions of the non-homologous chromosomes.

Another case of such a somatic segregation seems to be the occurrence of white and red stripes on the pink corolla of the species hybrid *Nicotiana tabacum* (white corolla) × *N. Sanderse* (red corolla). Most frequently single white (1) or red (2) stripes have been found on the background of pink corolla but in a few cases I found a red and a white stripe (3) laying together on the background of pink. This somatic segregation can be also alternatively interpreted by interchange or by somatic non-disjunction but since they occurred too often and since studying the chromosome number in the pollen-mother cells (in about 250 cells of 10 floral buds) no cell with non-disjunction was found, it seemed more probable that somatic interchange between non-homologous (or partially homologous) chromosomes accounts for these somatic segregations (Fig. 4). If gene R is responsible for the red colour, being located in chromosome A, i.e., AR, after a chromatid crossing-over between chromosome AR and BO (AR + BO = pink), AR + AR = red, BO + BO = white), new chromosomes AO and BR should be formed. If cell with AR + BR chromosomes is capable to divide, red stripes should be formed; if cell with AO + BO is capable to divide further and reproduces
tissue—white stripes should be formed and, finally when both these kinds of cells divide, white and red stripes (adjacent) on the background of pink should originate. All these three kinds of stripes were observed. The variegations in the above-mentioned Nicotiana hybrids can be better interpreted by assuming interchange than non-disjunction for the following reasons. Cells with an interchange that do not lead to formation of acentric and bcentric chromatids have whole genoms of N. tabacum and N. Sanderæ while those with non-disjunctions should have one whole chromosome in excess or one chromosome lacking; consequently the interchange cells should be viable and should be able to divide further and compete with the normal hybrid cells, while there is a very small probability that cells with "non-disjunctions" especially those that lack a chromosome would be able to compete with the normal cells. The formation of white and red areas suggest that the tempo of the cell division in these regions is like that in the pink normal regions.

The cytological investigations by Teleziński (1935) on "unstable race" of Petunia violacea "with mosaic flower patterns", showed that all of the plants of this race are structural heterozygotes (p. 233). He interpreted the "unstable" flower patterns by the hypothesis of "unstable genes". It seems to me that his data can be much better interpreted by somatic interchanges.

It is quite possible that the great majority of the cases described as variegations as due to unstable genes represent chromosome interchanges in soma and that this interchange probably proceeds in the heterochromatic
regions. It is very desirable, of course, an arrangement of special experiments for verification of these suggestions.

In *Drosophila*, for example, Kaufmann (1934) observed configurations between the chromosomes in somatic cells that remind of chiasma formations in meiosis before genetic data were available upon the somatic crossing-over. The attention should be called here to the observations in *Drosophila* reported by Stern (1936) on the basis of which he stated that "Somatic crossing-over between X-chromosomes heterozygous for \( bb \) of inversion occurs within the inversion. It leads to a chromatid which possesses no fibre point and is thus eliminated, and to a complementary chromatid with two fibre points. This chromatid becomes fragmented and each fragment is included in a daughter nucleus" (pp. 727–28). It would be of great importance if such phenomena could be traced back to the exact point of crossing-over, *i.e.*, whether the somatic crossing-over has taken place in the active portion or in a small inert region. One could make a more general statement if in the excellent works by Schultz (1936) upon the variegation in *Drosophila* in relation to the inert chromosome regions and the increased numbers of Y-chromosomes, as well as in those by Stern (1936, 1936a) and Neuhaus (1936) upon the crossing-over between X- and Y-chromosomes in *Drosophila*, and in that by Sturtevant (1937) upon the preferential segregation in triplo-IV females of *Drosophila melanogaster* such points were stressed. It is clear now that the majority of the data are in agreement with our postulates (some of them will be mentioned later), while some might not be for various reasons.

Somatic crossing over, when it occurs in species hybrids with asyndesis, is of a great significance from an evolutionary as well as from a practical point of view. *Secale* species as well as *Triticum* species have heterochromatic regions (Kostoff, Dogadkina and Tihanova, 1935; Kostoff, unpublished). The investigations by Lebedeff (1932), Müntzing (1935), Wakar (in press) and Kostoff (unpublished) upon the proceeding of the meiotic processes in *Triticum × Secale* hybrids, especially upon the hybrids *Triticum vulgare* \((n = 21) \times Secale cereale (n = 7)\) showed that in some of these hybrids as a rule only 28 univalents appear, while in others (depending on the variety used from *Tr. vulgare*) one, two, three and sometimes even more bivalents are formed. This phenomena was explained before, that in the hybrids with bivalents an autosyndesis takes place during the meiosis between the chromosomes most probably from B and C (or as the Japanese school calls it D) genomes. In the light of the present studies it seems possible that in the hybrids with bivalents a somatic crossing-over between non-homologous
chromosomes has taken place, thus new chromosomes are formed, which have homologous segments. Such chromosomes can conjugate and form chiasmata. It is probable that the differences in the rate of somatic crossing-over that might be regulated by gene or genes like the Miniature in *Drosophila* (Stern, 1936) account for the differences in the behaviour of these species. It is also probable that somatic crossing-over should occur in heterochromatic regions or near them as it does in *Drosophila*. The chromosome conjugation during the meiosis as a result of somatic crossing-over can be easily explained in the following way: If *Secale* chromosome A with segments *a, h, b, c, d* conjugates with *Triticum* chromosome N with *n, h, m, p, q*...segments, segment *h* being a heterochromatic one, and if interchange takes place in chromatid stage (as it seems to be so) the following four chromosomes should be formed *a h b c d, a h m p q, n h b c d* and *n h m p q*. Two of these chromosomes should go into one daughter cell and the other two into the other daughter cell namely: *a h b c d* and *n h b c d* to one pole and the other two to the other pole or *a h b c d* and *n h m p q* to the one pole and *a h m p q* and *n h b c d* to the other. If *a h b c d* goes together with *n h b c d* the segments *h b c d* are homologous and should conjugate during the meiosis. The same behaviour should have the chromosomes of the other daughter cell. Such plants, no doubt, should be chimeras. Another alternative interpretation should be that different varieties of *Triticum vulgare* have unequal heterochromatic regions in the karyotype, those with more, or with larger heterochromatic regions should form more bivalents. Species hybrids with asynthetic meiosis cannot be used for transferring of the characters from a species on the background of another one unless interchange (somatic or gametic) between non-homologous chromosomes takes place. This can be expected to occur most probably between the heterochromatic segments. Its frequency can be increased by external factors that increase the frequency of interchange (X-rays, radium, temperature, etc.), or in using varieties that have genes which act in a similar way gene Miniature acts in *Drosophila* as shown by Stern (1936).

The most effective agents which we know that increase the frequency of somatic chromosome interchanges seem to be X-rays (radium acts similarly) and temperature. The effects of the X-rays upon the chromosome interchanges is more thoroughly studied, therefore I shall consider here these data more extensively.

It is generally accepted now by a large number of the cytogeneticists that X-rays induce chromosome "breakage" and that the "broken" ends joint. Such is thought to be the mechanism of chromosome interchanges.
The cytological observations at various intervals of time after X-raying plants or animals show consistently that chromosome fragments are formed as a result of X-raying. It seems to me, however, that a large number of the fragments are results of chromosome interchanges (between homologous or non-homologous) and that the majority of the chromosome dislocations result from chromosome interchange between homologous or non-homologous chromosomes, in a similar way a crossing-over proceeds during the meiosis between "homologous" chromosome parts. The principal question that arises here would be: how X-rays do act upon the chromosomes and their milieu biochemically and biophysically?

Our knowledge at the present time does not suffice to answer definitely this question, but some observations made upon this problem would help greatly for advancing the most probable working hypothesis. The important statement by J. Clark (1937, Science, No. 2229), that X-rays induce denaturations in the proteins regardless of alcalinity or acidity of the solution and of the temperature of which the experiment is performed, is of great significance for our discussion. In respect to the biophysical changes Seckt (1901) and Williams (1925) found that short exposure of various plants to X-rays increases the rate of the cytoplasmic streaming and Brownian movement, while longer exposure retards them. Radium rays have the same effect (Zuelzer and Philipp, 1925; Williams, 1925). Certain intensity and duration of exposure to X-rays cause an increase in the cytoplasmic viscosity (Seckt, 1902; Williams, 1923). The chromosomes are protein bodies and would react to the X-rays as proteins do react. High temperature increases also the cytoplasmic viscosity (Heilbrunn, 1924, 1928) and at a higher and longer exposure the proteins coagulate. Both these agents induce chromosome associations in somatic cells. The studies carried out by Marquardt (1937) supply good evidence in this respect. He has induced chromosome conjugation in Bellevia romana during the pollen mitosis by X-ray irradiation as his figures 2 and 4(a) show. The pollen have haploid chromosome number, nevertheless X-ray treatment induces chromosome association in polyvalent groups, which means that the conjugations take place between non-homologous chromosomes. The drawings by Marquardt can be interpreted as conjugations between regions near the centromeres and distal ends; where the heterochromatic regions are usually located. Stone (1933) and Mather and Stone (1933) in studying the effect of X-rays on Crocus and Tulipa root tips, found that cells which actively divide are unaffected by irradiation. Marquardt (1937) came to similar conclusions, stating "dass in der Mitose vorhandene Translocationen nur während des Ruhestadions erfolgen" (p. 151). These observations, supporting
Belling's (1933) and Darlington's (1932-37) theory of the chromosome division (or reproduction) during the prophase, suggest that Belling's theory (1933) of crossing-over during meiotic prophase, might be as well applied for these interchanges with great probability. I shall consider here, however, another alternative interpretation, since it seems to me that all observations, recorded by various authors, cannot be quite harmoniously unified. In studying the time of chromosome "breakage" in *Drosophila* Patterson (1935) concluded that breakage is not delayed even for a single cell generation (p. 241). Lewitsky and Araratian (1931) are inclined to interpret their results from the X-irradiation experiments in assuming "a marked postaction" of the X-rays in form of a durable chemical change in the plasm or chromosomes. Patterson's statements upon this question are somewhat contradictory. He (1933) crossed X-rayed virgin *Drosophilas* with untreated males. In studying the mosaic flies he concluded "that their male parts lose the paternal X with the same frequency as the maternal X. This leads to the suggestion that the effect of radiation on the elimination of the X-chromosome is in part indirect, probably operating through the cytoplasm" (p. 51). In a more recent publication, Patterson (1935) drew the following conclusion: "We have been unable to find any evidence in support of this view that the effect of the irradiation may be indirect by the way of the cytoplasm" (p. 241).

Mather (1934) X-rayed *Vicia Faba* and *Tradescantia* and found "fragments" in the divisions occurring soon after treatment. The "fragments" in *Vicia Faba* appeared at late metaphase and anaphase while in *Tradescantia* before the metaphase. These differences he ascribed to the differences of forces that condition terminalization of chiasmata, *Vicia* having a much smaller terminalization coefficient. Riley (1936) found fragments at metaphase and anaphase of the microspore division within one hour after irradiation of *Tradescantia* buds and concluded that they should be "broken off" during the metaphase or late prophase. Riley (1936) reported that clumping of the chromosomes, treated during the meiosis and examined at the meiotic metaphase, was the only physiological effect he was able to notice. In his figures (especially fig. 12) drawn from the first pollen metaphase, chromatid exchange is shown between non-homologous chromosomes. The chromosomes are differentiated in length, the proteins of various chromosomes and genonemes have different isoelectric points and probably coagulate (undergo denaturation, J. Clark, 1937) at various periods of time depending on the intensity and on the duration of exposure. The discontinuation of
the chromosomes probably occurs at the place where the proteins of the chromomeres coagulate partially or completely. Identical chromomeres have the same kind of proteins and should undergo changes at the same time, while the proteins of similar chromomeres should coagulate at about the same time in homogenous milieu. In the active regions "homologous" chromomeres (i.e., equal) should coagulate at about the same time, consequently in more active segments chromatid discontinuation should occur at about the same time. But this does not always seem to occur, probably, because the chromosome milieu is very heterogeneous and the intensity of action of X-rays is not equal at any point. Some chromatid parts might join again at the place of discontinuation or with some of the ends of the chromatids from other chromosomes that are in proximity. Thus the effect of X-rays on the chromosome dislocations seems to be in two ways: (1) They induce association between chromosomes, often an association in the heterochromatic regions of the homologous or non-homologous chromosomes and the latter exchange parts. (2) They induce chromatid discontinuation, denaturing some proteins of certain chromomeres or in between them. (This process is known in the literature under the term "breakage".) Chromatid fragments of the same chromosome might join again with each other or with some fragments of chromatids of other chromosomes. The latter might occur more readily when X-rays induce conjugation between non-homologous chromosomes (point 1). The drawings given by Riley (1936) and Marquardt (1937) seem to fit quite well to the above advanced interpretations. It seems plausible to assume that the chromosomes of the heterochromatic regions of all chromosomes have closely alike proteins and they get denaturated at about the same time under the activity of X-rays, consequently, the discontinuations and junctions anew between non-homologous chromatids in these regions are to be expected more frequently. In other words, chromosome interchanges following X-ray treatments should occur most frequently in the heterochromatic regions.

The observations which showed that X-ray irradiations induce conjugation between non-homologous chromosomes suggest at the same time that the same agent upsets the force that prevents a coalescence of the like or similar parts of the chromosomes in the somatic cells. If the material per unit length of the heterochromatic regions of non-homologous chromosomes is less heterogeneous than the material of the euchromatic regions of two closely situated chromosomes, as it should logically be so, the coalescence should then more readily occur in the heterochromatic regions.
Some arguments that support the above given postulates will be later mentioned. Now I shall point out to the theories that attempt to interpret the mechanism of crossing-over and chromosome interchanges. I shall mention here the theory advanced by Serebrovsky (1929) who postulated a hypothetical tendency for chromosomes to become attached and subsequently "break" apart at different points. On this hypothesis, X-rays should increase the tendency for the chromosomes to become attached, or as we said above "coalesced". When this theory was advanced no cytological evidence was known in favour of such an attachment. I mentioned above that such observations were recently reported. It might be added here, that there seems to be a preferential chromosome attachment in the heterochromatic regions. This interpretation can be assumed for the cases when an attachment first occurs and then a discontinuation ("breakage"), it cannot explain, however the cases when first discontinuation and then reunion takes place. Patterson, Stone and Suche (1934) believed, "that most translocations, though not necessarily all, occur by an attachment followed by—breakage—" (p. 368). Muller called the broken-ends "sticky", consequently they should reunite. Stadler (1932) pointed out that simple translocations and terminal inversions have never been shown to follow irradiation and advanced the hypothesis that true ends cannot rejoin having a specific non-fusibility. Catcheside (1935, 1936), on the other hand, explained the reciprocity of structural changes without assuming a specific non-fusibility of the ends. He inferred that the reciprocity of changes is due to the high unlikelihood of an end of one chromosome lying near the point of discontinuation of another (cf. Darlington, 1937). The procedure of chromosome interchanges seems to be similar in many respects to that of the crossing-over. There are two hypotheses that attempt to interpret the procedure of crossing-over namely Belling's hypothesis (1933) and Darlington's hypothesis (1937). Belling postulated formation of new attachments between the newly formed chromomeres during the early prophase when the chromonemata of two homologous chromosomes associate. There are two serious arguments against this hypothesis: (1) The formation of new chromomeres, i.e., the reproduction of the chromonemata does not seem to proceed at that stage since the chromosomes consist of two chromatids during the anaphase (Marshack, 1937; Atwood, 1937; Gates, 1937; etc.) and (2) on the basis of such a hypothesis very often acentric and bicentric chromatids should be formed unless special probable forces are postulated that regulate the junctions between newly formed chromomeres and disjunctions between the old ones. Darlington (1935, 1937), on the other hand,
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assumes "breakage" in two of the four chromatids under the force of torsions. "The two broken ends will twist round their unbroken sister chromatids, thus releasing the coiling of the two which determined the breakage".—"The broken chromatids will reunite when, in the course of uncoiling, one of their ends first meets another. This will always be the end of a chromatid of a partner chromosome. Crossing-over will have occurred, and when the lapse of attractions leads to separation, a chiasma will appear" (p. 550). Darlington's hypothesis considers definite forces that are probably involved during the early prophase in connection with the spiralization and uncoiling of the chromosomes and chromatids as well as the chromosome attraction and repulsion. The weak point of Darlington's hypothesis is the postulate of chromatid "breakage," "uncoiling" and then "reunion". It seems very doubtful that such a "mechanism" will work so perfectly when the "axes" and "wheels" are built up from "colloid material" like that of the chromosomes. If we assume first a "breakage", i.e., discontinuation and then twisting, it seems very doubtful that the "broken" chromatids should always reunite, and that the reunion should always occur between partner chromatids. If Darlington's hypothesis was correct we should find very often fragments during the late metaphase and anaphase resulting from breakage but failure of reunion as we often find in irradiated material. The first objection to Belling's hypothesis is also an objection to Darlington's hypothesis. The intimate procedure of crossing-over as well as the chromosome conjugations and interchange of parts can be more satisfactorily interpreted when special studies are undertaken in the light of researches in the colloid chemistry in connection with the coalescence and miscibility of liquids. It seems that such a kind of research would throw light on the nature of the preferential interchange in the heterochromatic regions too.

I shall point out here to a striking correlation rather than coincidence between the position of the heterochromatic regions and the place of interchange following X-ray irradiation and temperature treatments. This phenomenon seems to occur in nature too. Preparations of root tips of Crepis capillaris fixed in chrom-formol fixatives, stained in iron-hæmotoxylin, and somewhat overdistained showed a differential staining, the proximal parts being darker and often small portions of the distal ones (Fig. 5). Similar differential stainings can be seen on the microphotographs reported by Matsuura (1937, his figs. 1 and 2). When one compares the chromosome interchanges in Crepis capillaris induced by X-ray irradiation and reported by Lewitsky, Shepeleva and Titova (1934) as well as the new karyotypes that
might be derived from such having A-, C- and D-chromosomes and compared with the karyotypes found in nature and reported by Babcock (1936), one gets the impression that the majority of the newly originating

karyotypes resulted from chromosome interchanges in the heterochromatic regions (distal, proximal, or both) of non-homologous or homologous chromosomes, or finally—resulting from interchanges involving only one chromosome, i.e., following "fold backs". In Fig. 6 it is shown diagrammatically how an interchange might take place in the proximal regions involving one chromatid of A- and one of D-chromosomes. This way new chromosomes have obviously resulted, one with two large arms ($a_1$ $d_1$) and another with
**Fig. 6.**

small arms ($a_2 d_2-$, Fig. 6) in Lewitsky's experiments (1934, his fig. 4). In the other diagrams (Figs. 7, 8, 9, 10) I have drawn only the chromosomes

**Fig. 7.**

and the new types that would originate from interchange ($c - o$) in the heterochromatic regions. When interchanges occur in the regions as shown in Fig. 7 acentric and bicentric chromatids and chromosomes will be formed, both being not adapted to survive, the former, because it has not a centromere that regulates its normal behaviour, the latter, because it has two centromeres one often pulling to the one pole while the other to the other pole. Cells with discontinued bicentric chromatids can sometimes survive and give rise to new individuals and even new karyotypes (Stern, 1936; Kostoff, unpublished). Sometimes whole chromosomes can originate consisting chiefly from heterochromatic material (Fig. 8, $m, m_1$). This suggests the

**Fig. 8.**

idea that Y-chromosome in *Drosophila melanogaster* probably consists chiefly of inert material of X-chromosome. The production of a five-chromosome race of *Drosophila melanogaster* by Schultz (Morgan, Bridges and Schultz, 1935) is in favour of this idea. From interchange in the proximal heterochromatic regions of A- and C-chromosomes in *Crepis* (Fig. 8) two
new chromosomes could be formed $b$ and $n$. A chromosome pair like "'b'" occurs in the species *Crepis bungei*, chromosome pair like "'n'" can be found in *Crepis nicansis* and *C. albida*. If an interchange takes place further in the heterochromatic regions as between "'n'" and "'A'"-chromosomes as shown in Fig. 8, two new chromosomes: "'m'" and "'st'" can be formed. One chromosome pair, similar to chromosome "'m'", was found by Babcock in the species *Crepis montana* and *C. senecioides*; chromosome pair, morphologically similar to chromosome "'st'", was found in *Crepis Stoyanovii*. An interchange between chromosome C and A (fig. 8), when the distal ends are turned to a reciprocal direction, chromosome can be formed like "'m_{1}'", that is found in *Crepis montana* and like "'st'"-chromosome in *Crepis Stoyanovii*.

![Fig. 9](image)

When interchange takes place in the proximal regions between A- and D-chromosomes two possibilities are present: (1) When the distal chromosome ends point to quite opposite directions (Figs. 6 and 9 left), chromosomes "'i'" ($a_1 d_1$) and "'p'" ($a_2 d_2$) can be formed, type "'i'" being found in *Crepis incarnata, palestina, gymnopus* and *pulchra* and "'p'" in *Crepis parviflora* (comp. Babcock, 1936). (2) When the distal ends of the chromosomes point to and the same direction (Fig. 9, right) chromosomes "'D_{s}'" and "'b'" can be formed, "'D_{s}'" reminding of a pair of *Crepis setosa*; "'b'" of a pair of *Crepis bungei* and *incarnata* (cf. Babcock, 1936). The attempts made above to interpret the origin of some chromosomes is not an attempt to solve phylogenetic problems in the genus *Crepis*, since the reverse process is also possible, as for example, the formation of A- and B-chromosomes from "'i'" ($a_1 d_1$) and "'p'" ($a_2 d_2$) chromosomes, etc. "'Folding backs'" (Fig. 10) can also lead to chromosome reorganization. The above schemes as well as the results obtained by Lewitzky and his co-workers (1934) suggest that chromosomes with two arms are not necessarily more

![Fig. 10](image)
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primitive, from a phylogenetetic point of view, since they can originate at any time by interchanges.

Chromosome reorganizations, like those conditioned by X-ray irradiations were induced in Crepis by Shkvarnikov (1936) in applying abnormally high temperature. His fig. 2, in which some types of chromosome interchange in Crepis capillaris are given, suggests strongly preferential interchange in the heterochromatic regions.

Another factor that increase the interchange between the chromosomes is the aging. Heribert Nilsson (1931) noted that in Oenothera: "Mit dem Alter des Samens, also mit der herabgesetzten Keimfähigkeit, geht das Ansteigen des Mutationsprozents ausgesprochen parallel" (p. 326). Oenothera mutations are most frequently chromosome alterations. More recently Navashin (1933–36) and his co-workers have shown that with the aging of the seeds of Crepis and other plants the chromosomal "mutations" are significantly increased. The new type of chromosomes formed also remind of those formed under the activity of X-ray irradiations and those at high temperature. The formation of biceentric chromosomes following interchange most probably in the distal ends, as shown in our diagrams (Fig. 7) might account for the chromatin bridges observed by Navashin and Gerassimova (1935). Their drawings also suggest a preferential interchange between non-homologous chromosomes in the heterochromatic regions (N. and G., 1935, their figs. 18 and 19). Chromatin bridges were found by Barber (1938) during the second post meiotic mitosis in aged Paonia and Kniphofia pollen. He supposed, however, that bridges originate by union of the homologous ends of the two sister chromatids derived by division from one parent chromosome. Barber's interpretation is possible, but it seems to me that the possibility of interchange between non-homologous chromosomes is not excluded.

It was pointed out that X-rays induce denaturation of the proteins (Clark). The aging seems to be accompanied with similar biophysical changes. The works by Růžička (1922 and later) and his students showed that aging is accompanied with changes in the isoelectric points of the protein colloids which allows an easier denaturation (protoplasm-hysteresis).

In the light of the present discussion, the "mutations" of Oenothera that often occur can be better interpreted as interchanges in the heterochromatic regions of non-homologous chromosomes, aging being the factor that increase the rate of these interchanges.

In all cases of interchanges that are accompanied with protein denaturation it is possible that this phenomenon facilitates the coalescence of the chromatids and further interchanges when chromatid separation proceeds.
I shall finally mention of the interchanges that occur occasionally in centrifuged material. Centrifugal force transmits the chromosomes towards the centrifugal end of the cell and can mechanically facilitate a coalescence between some regions of non-homologous chromosomes.

The rôle of the heterochromatic regions can be evaluated in two ways from a phylogenetic point of view: (1) The heterochromatin might be the place where new genes originate. When they are totally inert a deficiency in these regions would not affect the vitality as well as the hereditary complex (cf. Kostoff, 1938a; Demerec and Hoover, 1936). By interchanges the heterochromatic regions can be transmitted from the proximal at the distal ends. (2) Interchanges in the heterochromatic regions lead to duplications of segments (active as well as inert). The evolutionary significance of the duplications was stressed recently by Morgan, Bridges and Schultz (1935). They supplied evidence which "confirms the hypothesis that the normal evolutionary increase in number of genes has been by 'duplication' of the previously existing genes, followed by diversifying mutation" (p. 287). Reduplications were found by many authors (Bridges, 1935; Kossikov, 1936; Offermann, 1936; Grünberg, 1937; etc.) and most of them share this opinion.

Closing the discussion I shall call once more the attention to the most probable origin of the atypical growth in plants and animals. The cancerogenic agents as methylcholontrene (Hearne, 1936) and Bacterium tumefaciens (Winge, 1927) induce chromosome figures that remind us of chromosome association during the prophase. Such a chromosome behaviour obviously leads to somatic crossing-over and interchange between non-homologous chromosomes. These phenomena were found in animals as well as in plants (Stern, 1936; Jones, 1937; etc.). I showed above that chromosome alterations, most probably interchanges in species hybrids, occur quite often. Species hybrids like Nicotiana glauca × Langsdorffii, N. paniculata × Langsdorffii, N. rustica × Cavanilliesii, N. glauca × longiflora, etc. form large tumors (Kostoff, 1935, 1937, etc.). The chromosome number in the tumors is usually equal to the somatic chromosome number and rarely chromosome duplications and aneuploidy has been found. Hence it seems most probable that atypical growth results from interchanges between non-homologous chromosomes and it probably occurs preferentially in the heterochromatic regions.

Discussing the data upon the heterochromatin and its behaviour recorded by various workers and adding some new observations into this line of work I attempted to correlate a series of phenomena connected with chromosome
interchanges and crossing-over. In doing this a number of the existing theories and hypotheses were criticised in the light of new researches. For a series of phenomena new interpretations were given. Some of them should be considered at the present time as working hypotheses, others having more solid experimental background can be treated as theories that generalize the knowledge at the present time in the respective fields of research.

Extreme empiricists do not often evaluate correctly the significance of the theories which mobilize the present knowledge in a certain field of the biological sciences and stimulate research into most promising directions.

As a controversion to the strict empiristic conceptions in experimental biology I shall quote Willstätter's opinion which seems to express much better the value of hypotheses in the experimental research work than a sceptical denial of any stimulating ideas which might lead to a disarmament to a standstill. Willstätter wrote: "It is not important for the scientist whether his own theory proves the right one in the end. Our experiments are not carried out to decide whether we are right, not to prove that we are right, but to gain new knowledge. It is for knowledge's sake that we plow and sow. It is not inglorious at all to have erred in theories and hypotheses. Our hypotheses are intended for the present rather than the future. They are indispensable for us in the explanation of the secured facts to enliven and to mobilize them and above all, to blaze a trial into unknown regions towards new discoveries." But there is no doubt, that it is the experiment which finally decides; it proves or disproves the theory and hypothesis.

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DESCRIPTION OF FIGURES.

Fig. 1.—Microphotographs from two cells at several levels from the root tips of Triticum timopheez during the mitosis. Roots fixed in fixation platine chlorid and commercial formalin fixation and stained in gentian violet. Five pictures at the right side are taken with lower magnification than the pictures at the left. Note: (1) Chromonemata consist of: (a) chromomeres that stain; and (b) substances in between that do not stain. (2) In some regions (usually distal and proximal) the chromomeres are darker and more closely situated.

Fig. 2.—Several chromosomes drawn separately from one and the same metaphase plate from Triticum monococcum (It is aimed to show the lighter and the darker regions). Note that the distal ends stain darker.

Fig. 3.—Late anaphase from a tapetum cell of the hybrid Nicotiana bauriensis X N. Sanderrae. Note two chromatin bridges representing bicentric chromatids.

Fig. 4.—Parts from the flowers (corollas) of the hybrid Nicotiana tabacum X N. Sanderrae. Note: (1) Left—Black (red) stripes (AR, BR) on the punctured (pink) background (AR, BO); (2) In the middle—White stripe (AO, BO) on the punctured (pink) background (AR, BO); and (3) Right—Black (red) and white stripes on the punctured (pink) background. Down—diagram showing the probable origin of the stripes as a result of a chromatid crossing-over in soma (corollas) between A-chromosomes carrying the gene for red (R) and B-Chromosome (cf. text).

Fig. 5.—Differential staining of the chromosomes in Crepis capillaris after Lewitzky’s chromformol fixation and Haematoxylin staining.

Figs. 6, 7, 8, 9 and 10.—Diagrams showing the mode of origin of new chromosomes following chromatid interchanges between non-homologous chromosomes in the heterochromatic regions (cf. text).